

Copenhagen

BODY SIZE AND METABOLIC RATE IN THE ANIMAL KINGDOM

BY

ERIK ZEUTHEN

H. HAGERUP - COPENHAGEN

1947

LANE LIBRARY
SAN FRANCISCO



Digitized by the Internet Archive
in 2026

BODY SIZE AND METABOLIC RATE IN THE
ANIMAL KINGDOM

BODY SIZE AND METABOLIC RATE IN THE ANIMAL KINGDOM

WITH SPECIAL REGARD TO THE
MARINE MICRO-FAUNA

BY

ERIK ZEUTHEN

*Forsvaret finder Sted Tirsdag den 11. November 1947 Kl. 14
i Universitets-Annekset, Auditorium A, Studiestræde 6 o. G.*

H. HAGERUP - COPENHAGEN

1947

Denne Afhandling er af det matematisk-naturvidenskabelige Fakultet
antaget til offentlig at forsvares for den filosofiske Doktorgrad.

København, den 17. April 1947.

Niels Nielsen
h. a. dec.

Reprinted from
Compt.-rend. Lab. Carlsberg, Sér. chim., Vol. 26, N° 3.

BIANCO LUNOS BOGTRYKKERI

BODY SIZE AND METABOLIC RATE IN THE ANIMAL KINGDOM WITH SPECIAL REGARD TO THE MARINE MICRO-FAUNA

BY
ERIK ZEUTHEN

CONTENTS

	Page
Preface	20
1. Introduction	21
2. Technique. Biological factors influencing the measurements.....	24
2.1. Technique	24
2.11. Respiration measurements with the Cartesian diver of the μ l type	24
2.12. Respiration measurements with the Cartesian diver of the 0.1 μ l type.....	27
2.13. The technique of determining the temperature-metabolism curve.....	30
2.14. Oxygen determinations by Winkler titrations ad modum KROGH	32
2.15. Weighings.....	33
2.16. N-determinations of the order of γ	33
2.161. Modification of the receiver acid and the titration	35
2.162. Destruction.....	37
2.163. Control experiments and accuracy estimation	38
2.17. N-determinations of the order of mg	43
2.18. Determination of the respiration/N	43
2.2. Biological factors in the metabolism measurements	44
2.21. Temperature	45
2.22. Activity	45
2.23. Food ingestion	46
2.24. Light.....	46
2.25. Rhythms.....	46
2.26. Geographical latitude and season of the year.....	46
2.27. Oxygen pressure.....	47

	Page
2.28. Body size.....	47
2.29. Position of the animals in the zoological system.....	47
3. Experimental part	48
3.1. Special problems concerning the metabolism of the micro-fauna	48
3.11. Metabolism during natural activity variations	48
3.111. Echinodermata and Vermes	48
3.112. Mollusca.....	49
3.113. Crustacea.....	57
3.12. The effect of the oxygen pressure on the metabolism and the activity of some micro-animals.....	58
3.13. The effect of the temperature on the metabolism of the micro-fauna	65
3.14. Experiments on the floating and swimming of some micro-animals in the plankton.....	69
3.141.) Reference to informations given elsewhere in this	
3.142.) paper	71
3.143. Oxygen consumption of single larvae at different states of activity.....	71
3.144. The metabolism of the isolated velum as compared with the metabolism of the intact, swimming mollusc larva	73
3.145. Swimming with reduced metabolism owing to low temperature	76
3.146. Conclusion.....	78
4. Theoretical part	78
On the relation between body size of the animals and their metabolic rate.	
4.1. Introduction	78
4.2. Metabolism/N of marine animals in relation to their size (N-content) with special regard to the micro-fauna	81
4.21. Coelenterata.....	82
4.22. Echinodermata	82
4.23. Vermes	84
4.24. Mollusca	86
4.241. <i>Nassa reticulata</i>	89
4.242. <i>Littorina littorea</i>	90
4.243. <i>Mytilus edulis</i>	94
4.25. Crustacea	98
4.251. Experiments with Winkler titrations	98
4.252. Diver experiments.....	99
4.253. Morphology.....	101
4.254. Activity	101
4.255. Ontogenetic state of development.....	102
4.256. Living habit	102
4.257. Species, group	102
4.258. <i>Balanus</i>	103

4.26. Pisces	105
4.27. Conclusion	105
4.3. Metabolism/weight in relation to the body weight of the animals	105
4.31. Echinodermata	107
4.32. Vermes	107
4.33. Mollusca	109
4.34. Arthropoda	111
4.35. Pisces	115
4.36. Amphibia	117
4.37. Reptilia	119
4.38. Aves and Mammalia	121
4.39. Conclusion	124
4.4. Comparison and analysis of the body size-metabolic rate curves	
4.41. Metabolic intensities in the animal kingdom as compared on the basis of metabolism/N and metabolism/weight....	124
4.411. Comparison of the metabolism of animals on the basis of the metabolism/N value.....	124
4.412. Comparison of the metabolism of animals on the basis of the metabolism/weight value.....	127
4.42. The percentage decrease in metabolic rate at a ten times increase in body size.....	131
5. Discussion.....	134
5.1. Metabolism and food requirements.....	134
5.2. Metabolic rate and body size.....	135
5.21. Introduction.....	135
5.22. The author's own investigations.....	136
5.23. Metabolic rate variations throughout the life of the individual-animal. The possible bearing of this phenomenon on the general understanding of the metabolic rate — body size relations.....	140
5.24. Why does the metabolism per unit of weight or N decrease with increasing body size of the animals?.....	149
5.25. The mathematical description of the phenomena	152
5.3. Evolution, metabolism, and heat regulation.....	154
6. Summary	155
7. References	158

PREFACE

In 1943, Dr. GUNNAR THORSON invited the present author to perform respiration measurements on the marine micro-fauna, especially the larvae of marine bottom animals.

The reason for this invitation was, on the one hand, that Dr. THORSON considered such measurements valuable for the elucidation of his own ecological problems and, on the other hand, that I had participated already for some time in the work going on at the Carlsberg Laboratory on the development of new micro-chemical and micro-physiological methods. Thereby, I had gained some experience in the application of the Linderstrøm-Lang micro-respirometer, the Cartesian diver, which could be considered well suited for the mentioned purpose.

During the course of my work I have met with the utmost goodwill from all sides. Lektor GUNNAR THORSON, Ph. D., placed his small laboratory at Helsingør at my disposal during three summers (1943, 1944, and 1946) and supported my work in numerous different ways. Professor K. LINDERSTRØM-LANG, Ph. D., in whose department at the Carlsberg Laboratory the chemical analyses were performed, granted me valuable guidance and, moreover, he has most generously given Mrs. KAREN MARGRETHE GJERLØFF time to assist me in the nitrogen determinations according to the Carlsberg method.

My former chief, the head of the cytochemical department of the Carlsberg Laboratory, HEINZ HOLTER, Ph. D., placed all the laboratory facilities at my disposal and offered his time for a long series of instructive discussions which I shall hardly forget. My present chief, Professor P. BRANDT REHBERG, Ph. D., has offered me extremely good working conditions in the Laboratory of Zoo-physiology, University of Copenhagen.

In Helsingør, I was assisted by stud. mag. PER FORCHHAMMER, stud. real. EINAR P. ELIASSEN, and by my wife, Mrs. ELISABETH ZEUTHEN. The personnel of the State's Harbor Office in Helsingør has been very helpful to us. Miss GRETHE KRUGHØFFER has been of great help to me during the elaboration of the manuscript and so has my translator and colleague Miss HILDE LEVI, Ph. D.

I wish to express my sincere thanks to all my above mentioned colleagues for the instruction I received, for their help, and for their stimulating comradeship in the years of our collaboration.

Copenhagen, June 2nd, 1947.

I. INTRODUCTION

RUBNER, in 1883, observed that the metabolism of warm-blooded animals when expressed per kg of the body weight (metabolic rate) is the smaller the larger the animal but, on the other hand, is approximately constant when calculated per surface unit (RUBNER's surface law (98, 99)). Since then, the comparative study of the metabolic rate of animals has attracted the interest of physiologists and zoologists.

For animals of equal proportions it holds that the surface is proportional to $\sqrt[3]{W^2}$ (where W is the weight) or, in other words, proportional to $W^{0.67}$. While most investigators agree that the metabolic rate decreases with increasing size of the animals, they hold different views as to whether it varies as a function of the surface, *i. e.* of $W^{0.67}$, or whether another exponent should be adopted. The discussion thus deals mainly with the value of the exponent, but much confusion prevails in the recent literature. To quote KLEIBER (Ann. Rev. Physiol. VI, 1944):

"In ten papers (from eight laboratories) metabolic rates of rats are expressed per unit of the surface area. Four of the authors did not state how they measured or calculated this area. One multiplied the $2/3$ power of body weight by 7.42, another by 9.1, a third by 10, to calculate the surface area in square decimeters. One author multiplied the $3/5$ power of body weight by 12.44 and two have calculated a new surface-weight relationship, presumably by an intricate logarithmic interpolation between three older ones ($7.42 \cdot W^{2/3}$, $11.36 \cdot W^{2/3}$, $12.44 \cdot W^{3/5}$). The new formula thus derived, $0.001 W^{0.63}$, expresses the surface area of rats in square meters, when the weight (W) is given in grams. That—for rat metabolism only—is this year's result of a century of surface law.

Reducing the metabolic rate to a power function of body weight such as the $2/3$ or $3/4$ power is valuable and essential for some problems in comparative physiology. Every scientist also should, of course, be free to use any surfaces he likes, but the time is ripe for asking authors, who use their personal rat surfaces, to supplement their figures with data which make their results comparable to those of other workers. — — —"

The present work aims at an unprejudiced study of the relation between size and metabolic rate throughout the animal kingdom. The metabolic rate will simply be expressed as the O_2 -consumption

per wet weight or per N-content of the animals. When combining the values available in the literature with the author's own measurements we arrive at a size variation of the animals of c. 10^{12} times, corresponding to the difference in size between small marine plankton organisms and large mammals.

The literature comprises a great number of measurements on larger animals, while measurements on smaller animals are scarce, and measurements on micro-animals are lacking almost completely. Therefore, the experimental part of the present work deals with the study of small organisms, especially micro-animals. So far, it has only been possible, however, to investigate the metabolic rate of marine micro-animals.

Mainly owing to the insufficiency of the methods applied, our knowledge of the metabolic rate of marine micro-animals is still very limited. PÜTTER (93) assumed a general validity of RUBNER's surface law and he, therefore, considered it admissible on the basis of our knowledge of the respiration intensity of larger marine animals to calculate the respiration intensity even of small animals. As already emphasized by RUBNER (100), and as it will also appear from the present work, the results of these calculations are of a quite false order of magnitude, because RUBNER's law is not generally valid. Only few satisfactory measurements of the oxygen consumption of plankton organisms are available. SPÄRCK (101) determined the O_2 -consumption of oyster larvae by means of Winkler titrations and he drew the conclusion that the oxygen consumption of the larvae is much higher than that of the adults. An accurate comparison, however, was impossible because a determination of the respiring matter in the larvae could not be performed with reasonable accuracy at that time. From the Plymouth laboratory, measurements are available of the metabolic rate of the relatively large plankton copepod *Calanus finmarchicus* (MARSHALL and NICHOLLS (80)). The respiration intensities found for these animals appear to be somewhat high (2–3 times) as compared with the values found in the present work on other copepods. However, an exact comparison cannot be made, since the British authors only mention the average weight of one animal, while the present author determined the N-content of the single experimental animal.

The Cartesian diver (LINDERSTRØM-LANG (72)) proved to be well suited to the study of the oxygen consumption of marine micro-organisms. The principal difficulty was thereafter to develop

a method of measuring the quantity of material used in every single respiration experiment, in other words, a method of measuring the mass of the animal employed.

Primarily, it has been the aim of the present experiments to compare the metabolic rate of the micro-fauna with the numerous earlier determinations, performed by Danish investigators, of the metabolic rate of the bottom animals (SPÄRCK (103), THAMDRUP (107), THORSON (108)).

Such a comparison, however, involves that the respiration intensity of the micro-animals—in analogy with all previous Danish experiments on bottom animals—should be expressed as O_2 -consumption per wet weight of the animals, where the wet weight refers to the tissue of the animals and, thus, in the case of mollusc larvae does not comprise the heavy shell. It should be emphasized, however, that the wet weight appears to be unsuited as a basis of comparison, because the water content of the tissue may vary from one animal to the other. It would be more reasonable to calculate the O_2 -consumption per dry matter or, still better, per nitrogen of the tissues.

When the present experiments were planned (in the spring of 1943), a method for the determination of nitrogen in the small quantities of material normally used for respiration experiments in the Cartesian diver was under development at the Carlsberg Laboratory (BRÜEL, HOLTER, LINDERSTRØM-LANG, and ROZITS (15)). Since this work was in good progress, and since it could be regarded as certain that the method would work satisfactorily, the author decided to carry out a series of experiments on the respiration of the marine micro-animals by means of the Cartesian diver (LINDERSTRØM-LANG (72, 73, 75), HOLTER (41), LINDERSTRØM-LANG and HOLTER (74), ZEUTHEN (118)). The size of the animals should be expressed by their N-content determined with the micro-method mentioned (here to be called the Carlsberg method). In order to make it possible to compare these micro-experiments with previous studies on larger animals, new experiments on larger animals were planned; the results obtained should be calculated as respiration per N and as respiration per wet weight of the tissue. The respiration should be determined according to the Winkler method of KROGH (64) and the nitrogen by PARNAS-WAGNER's micro-method (88).

2. TECHNIQUE. BIOLOGICAL FACTORS INFLUENCING THE MEASUREMENTS

2.I. *Technique.*

2.II. Respiration Measurements with the Cartesian Diver of the μ l Type.

A detailed description of the technique of respiration measurements by means of the Cartesian μ l diver was given by HOLTER (41); hence, only a short outline will be presented here.

Fig. 1 shows the diver charged for an experiment. The diver is a small bottle with a long neck, a blown up bulb, and a

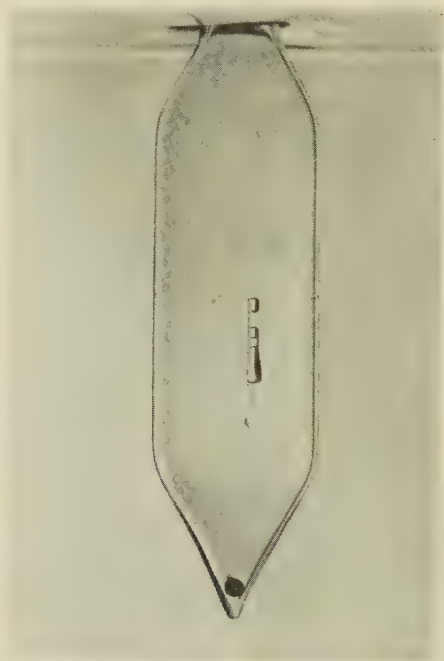


Fig. 1. The μ l-diver in floating position, (LINDERSTRØM-LANG (75)).

(The medium seal in the upper part of the neck is not well visible in this diver).

solid tail containing just so much glass that the charged diver will float in HOLTER's medium (a strong salt solution, specific gravity 1.326) without rising or falling. The animal or animals whose respiration has to be measured are placed in a drop of water situated in the

bulb of the diver. The neck is closed by means of several liquid seals. The lowest one consists of a weak alkali solution, isotonic with the drop in the bulb, the second one of paraffine oil and, finally, the liquid occupying the uppermost part of the diver neck is the strongly saline medium in which the diver floats. Each of the three liquid seals in the neck of the diver has its special function: the lowest one absorbs the carbon dioxide formed in the respiration process proceeding in the bottom drop; accordingly, the air inside the diver is always carbon dioxide free. Paraffine oil is a very poor solvent for water vapour and, therefore, an effective brake against loss of water by evaporation from the interior of the diver. The flotation medium in the opening of the neck effectively prevents air from diffusing in and out of the diver. This is possible because, owing to its high salinity, the flotation medium dissolves only c. $\frac{1}{10}$ of the quantity of air as does pure water. Since the CO_2 -tension inside the diver always equals zero, and since the water vapour tension of the air is always constant (in the upper air space of the diver governed by the vapour tension of the surrounding medium, in the two lower air spaces, however, by that of the bottom drop and the isotonic alkaline seal) the oxygen consumption of the animals is the only process involving a change of the quantity of air enclosed in the diver. The gas volume of the floating diver is constant, and the decrease in pressure necessary to secure this constancy is a measure of the quantity of oxygen consumed. This quantity can be calculated with sufficient accuracy from the equation

$$\frac{dV}{dt} = (p - p_1) \cdot V.$$

Here, $\frac{dV}{dt}$ is the oxygen consumption (μl) per unit of time, $p - p_1$ the change in pressure (in atmospheres) within the diver in the same time, and V is the total air space of the diver (μl) in the floating position (in the present experiments 7–16 μl).

The diver can only be kept floating in the flotation medium provided the equilibrium condition is fulfilled, *i. e.* if the average specific gravity of the charged diver is equal to the specific gravity of the flotation medium. This demands that the air space of the diver is of a definite magnitude relative to the weight of the glass and the various liquids in the diver. This condition can be approximated, but not finally be reached, by a precise charging of the diver,

adjusted in advance. After the charged diver has been transferred to the flotation medium, the magnitude of the air volume can be regulated more precisely by means of a liquid manometer, one branch of which opens into the flotation vessel. Thus, we can adjust and read the "equilibrium pressure" of the diver on the manometer. The equilibrium pressure is given in mm H₂O and is measured relative to the atmospheric pressure, as the other branch of the manometer opens to the atmosphere. In order to avoid corrections for fluctuations in the atmospheric pressure during the experiment proper, the latter branch of the manometer is not open to the free atmosphere, but to the constant atmosphere of a large bottle immersed into the thermostat (regulated to $\pm 0.01^\circ$). Immediately before the experiment is started, the bottle is opened to the air and again closed so that the pressure inside the bottle is equal to the atmospheric pressure at the start of the experiment.

Control experiments with empty divers (i. e. normally charged, however without respiring object) showed that the equilibrium pressure can be reproduced with an accuracy of $\pm 1/2$ mm H₂O; only if long experimental periods are applied, a marked change also of the equilibrium pressure of such control divers may be observed. In actual experiments, the effect to be measured should be so great ($> 1-2$ cm H₂O per hour) that the respiration can be calculated with an accuracy greater than ± 5 per cent. In very few experiments with weakly respiring animals we must reckon with a lower accuracy, *viz.* ± 10 per cent, of the respiration measurements. Mostly, the respiration intensities measured were of the order of $10^{-2}-10^{-1}$ μ l O₂ per hour; in some experiments with especially large divers, an oxygen consumption of up to $2-3$ μ l O₂ per hour was recorded.

The diver is charged by means of calibrated micro-pipettes of the braking type (HOLTER (41)). Firstly, the animal is pipetted into the bottom of the diver together with a well-known quantity of water; subsequently, the seals of NaOH and of paraffine oil are placed into the neck. The flotation medium drop is placed at the end of the neck and is then sucked into the neck by means of a very thin capillary pipette which is introduced into the diver. By means of this pipette so much air is sucked out of the diver that the medium enters the neck till the calculated distance.

2.12. Respiration Measurements with the Cartesian Diver of the 0.1μ Type.

A detailed description of the technique of measuring with the 0.1μ diver is given by ZEUTHEN (118). The diver is 10–20 times more sensitive than the μ diver and the oxygen consumption measured

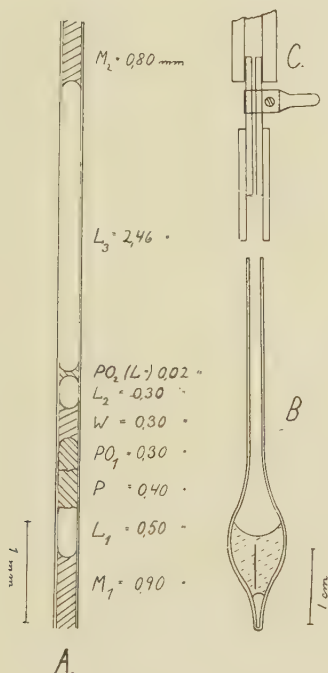


Fig. 2. The 0.1μ diver.

W: water containing the animal.

P: paraffine.

PO_1 , PO_2 : paraffine oil, prevents diffusion of water vapour.

M_1 , M_2 : flotation medium occupying both ends of the diver capillary; absorbs CO_2 and prevents the diffusion of other gases (ZEUTHEN (118)).

is generally of the order of $0.3 - 3 \cdot 10^{-3} \mu$ O_2 per hour. According to (42) and (118), the experimental error is about ± 10 per cent at the lowest, however considerably less at the higher respiration mentioned.

The 0.1μ diver is shown in Fig. 2. It is made from a glass capillary, 4–6 mm long and of a given low wall thickness. The

inner diameter (in the present experiments 0.28 mm) is only slightly larger than the diameter of the organism which is placed in the droplet (W).

Loss of water vapour from W to the surrounding medium is restricted because of the paraffine seals PO_1 and PO_2 . The solid paraffine P only serves the fixation of the drops in the interior of the diver. Due to the low solubility of H_2O in paraffine oil, however, PO_2 is very effective in preventing H_2O (as vapour) to be lost from the inside of the diver. CO_2 , on the other hand, dissolves readily in paraffine oil and, in fact, PO_2 which is very thin does not to any essential degree affect the absorption in the surrounding medium (M_2) of the respiratory CO_2 formed in W . The surrounding medium (c. $n/10$ NaOH in HOLTER's flotation medium) enters the diver in lengths represented by M_1 and M_2 and serves the reduction of the exchange of air by diffusion between the diver and the surrounding medium.

The diver floats in the alkaline flotation medium in such a way that the ends of the diver are only at c. $1/2$ mm distance from the upper and the lower surface of the medium (cf. Fig. 2). The saturation pressure of the air at the surface of the medium, *i. e.* at both ends of the diver, therefore decreases during the experimental period almost parallelly with the pressure in the interior of the diver. Thus, there is never any considerable difference in pressure according to which the air may diffuse into or out of the diver.

The charging of the 0.1 μ l diver requires a somewhat more complicated technique than that described previously for the μ l diver. By means of a small rubber plug the diver is placed at the point of a braking pipette. The animal is sucked in through the free end of the diver together with a drop of water. PO_1 is then sucked in and the point of the diver is pressed against a thin layer of solid paraffine, whereby P appears as a stopper at the point of the diver. $W + PO_1 + P$ are moved a little further inward the diver by means of a small glass rod. PO_2 is placed by means of a ball-tipped pipette (cf. 41) and, finally, the diver is immersed into the flotation medium and so much air is sucked out of the diver that the medium is sucked into the diver at the lengths M_1 and M_2 calculated in advance.

The measurements with the 0.1 μ l diver are performed according to the same general principles as discussed previously in the case of the μ l diver; thermostat, manometer, and measuring microscope are used in the same way. For special technical details and theoretical

considerations, the reader may be referred to the above mentioned publications.

An experiment for which 10–20 animals would be needed in the μl diver can be performed on one single animal with the same accuracy in the $0.1\ \mu\text{l}$ diver. This is a great advantage, for instance, when we have to do with mollusc veliger larvae. What we want to measure is the metabolic rate of the swimming larvae; but in experiments with the μl diver it will very rarely—and for certain species never—occur that all or a reasonable number of the animals contained in the diver swim simultaneously. If, however, the experiments are carried out on one larva in a $0.1\ \mu\text{l}$ diver, the metabolic rate of this larva can be measured as a function of its arbitrarily varying activity which can be observed in the microscope.

In the present work, the $0.1\ \mu\text{l}$ diver was also used for measuring the respiration as a function of the oxygen pressure. The measurements were made on one and the same microorganism which, as a consequence of its respiration, brings about the desired decrease in oxygen pressure in the diver.

The air space of the μl diver (c. $10\ \mu\text{l}$) is about 100 times greater than that of the $0.1\ \mu\text{l}$ diver (c. $0.1\ \mu\text{l}$). Nevertheless, the latter is only about 10–20 times more accurate than the former when applied to absolute respiration measurements. This is due to the fact that the $0.1\ \mu\text{l}$ diver charged without experimental animal in the water drop ("control diver") shows a much lesser constancy of the equilibrium pressure than the μl diver; a discussion of this phenomenon is given in (118).

The technical possibilities and the sources of error involved in the method are thoroughly discussed in (118) and (42). HOLTER and ZEUTHEN (42) have studied the problem to which extent the oxygen consumption of two as far as possible equal biological objects will deviate when the respiration is compared in diver measurements. Eggs and embryos of the ascidian *Ciona intestinalis* were used in these experiments, and it was found that the oxygen consumption of two fertilized or unfertilized eggs from the same female did not deviate much more (*i. e.* not > 20 per cent) than did their volumina (*i. e.* not > 16 per cent). Thus, according to our experience, differences > 20 per cent between the respiration intensities of two objects as found with the $0.1\ \mu\text{l}$ diver must be considered rather reliable and open to biological interpretation.

2.13. The Technique of Determining the Temperature-Metabolism Curve.

Experiments were carried out both with 0.1 μ l and μ l divers. The regulation of the thermostat was switched off and the temperature was changed in steps either by arbitrarily cooling with ice or heating electrically. However, temperature variations should not occur too rapidly, because rapid shifts in temperature induce creeping of the seals in the neck of the diver so that sodium hydroxide, oil, and medium come more or less into contact. The degree of creeping is essentially reduced when the temperature variations occur slowly.

The thermostat is made of wood and provided with a glass window only in front of the experimental vessels. Therefore, the temperature remained rather constant for a sufficiently long time at the level to which it had been adjusted.

After the desired temperature was reached, about 15 minutes elapsed before the measurements were started in order to secure complete temperature equilibrium between the thermostat and the immersed flotation vessels. During the experimental period the temperature obviously was not quite constant, but changed gradually by 0.2–0.3°. At the lowest temperatures applied (2–5° C.) the temperature variations could only be held within these limits when small pieces of ice were present in the thermostat during the whole period.

In all experimental series described here, 1–2 control divers were used, *i. e.* divers charged for an experiment, however lacking the animal in the water drop. The equilibrium pressure of these divers is constant at constant temperature and it changes slightly with varying temperatures in the thermostat. Thus, the equilibrium pressure of the controls was not quite constant, the changes, however, being only slight and always parallel for the two controls. Changes in the equilibrium pressure of the experimental divers are corrected for by means of the controls. A series of measurements is represented in Fig. 3. The ordinate shows the change of the equilibrium pressure throughout the experimental period, both for the control and for the experimental divers. The time is plotted as abscissa. It appears that the change of the equilibrium pressure in the respiration experiment minus that in the control experiment (= respiration effect expressed in cm H₂O per hour) can be calculated with a reasonable accuracy in all cases, although the percentage uncertainty

of the measurements performed at low temperatures is rather high. The "respiration effects" thus obtained are comparable only after conversion in the ordinary way to one temperature.

The gas volume of a charged diver depends markedly on the temperature in the thermostat owing, partly to the expansion of

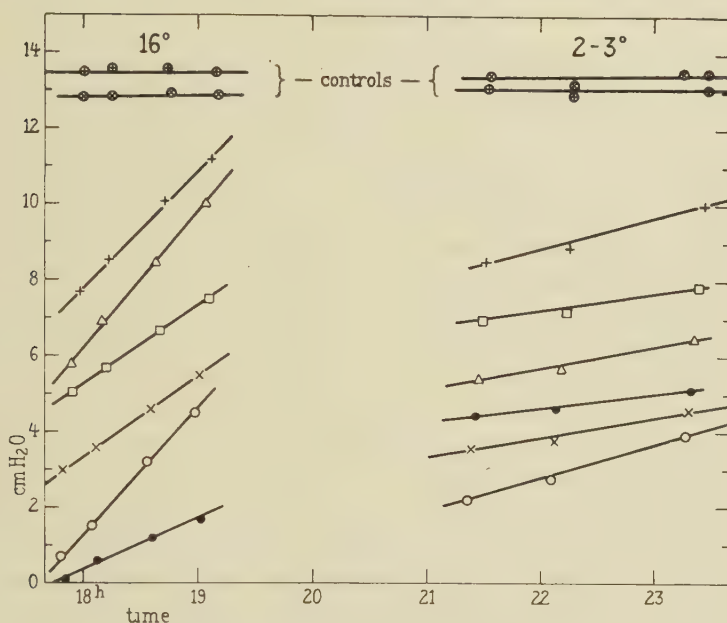


Fig. 3. Experiments carried out at different temperatures (μ l divers).

The figure shows the change in equilibrium pressure of two control divers (divers without animals) and of six divers charged with marine micro-animals. The experiments have been carried out at 16° C and at 2-3° C. We find rather well defined changes in equilibrium pressures at both temperatures.

the air, partly to the temperature dependence of the vapour tension inside the diver. Actually, the equilibrium pressure of the diver was not measured relative to the atmosphere, but relative to the pressure in a closed container immersed into the thermostat; obviously, the pressure in the container is a function of the thermostat temperature in (approximately) the same way as is the equilibrium pressure of the diver. Due to this fact, the equilibrium pressure in the diver did not change more than appears from Fig. 3 and, at all temperatures, the pressure could be read on the manometer. However, the change in pressure in the container and in the diver is only approximately

the same, because the vapour tension in the container (unknown humidity) and in the interior of the diver with its different air spaces is not the same.

2.14. Oxygen Determinations by Winkler Titrations *ad modum* KROGH.

The Cartesian diver is unsuited for respiration measurements on larger animals. However, a few experiments with the adult worm *Polydora*, weighing c. 2 mg, or with very small crustaceans (*Palaemon*, *Gammarus*, Isopods), weighing up to 5 mg, were performed in exceptionally large divers (50–170 μ l). For larger animals, KROGH's (64) Winkler method, based upon the syringe principle, has always been used. The animal or the animals are enclosed in a bottle of known volume, and the rate of decrease in the quantity of dissolved oxygen is determined by titration. By reasonably choosing bottles of different sizes and by varying the number of experimental animals, it has been possible to measure the metabolism of animals varying in weight from a few milligrams to c. 10 g. Still larger animals are not included in the present investigation, mainly because they were not easily available at Helsingør. There is, however, no difficulty in principle in applying the Winkler method also to such animals. By a combination of the diver method and the Winkler titration method, it ought to be possible in principle to include all marine animals (with special exceptions, of course) from the largest to the smallest, in the planned comparative investigation.

Generally, a few hours elapsed between the collection of the animals and the start of the experiment. The experiment began with an initial period of c. one hour's duration. During that period, the animals were kept at 16° C in the bottle, which was half filled with water, in order that they might become accustomed to the experimental conditions. Subsequently, most of the water was poured off and the bottle was filled with new water from a large container. The inlet-tube was introduced till the bottom and a quantity of water corresponding to about 3 times the volume of the bottle was led through. The start of the experiment was reckoned from the moment when the ground stopper was set in. During filling of the bottle it was attempted to disturb the animals as little as possible.

The control bottles containing water, however without animals, were treated analogously. After the lapse of c. 1–3 hours, 10 ml

samples were drawn with the aid of a Krogh syringe, both from the controls and from the experimental bottles, and the oxygen content of the samples was determined according to KROGH (64), however using one single modification. After addition of the reagents the bottles were shaken and after sedimentation ($1/2-1/4$ hour) they were shaken once more and were kept standing for sedimentation for another $1 1/2$ hours. Only when using this procedure, the high accuracy stated by KROGH was obtained. Since only respirations resulting in small reductions of the oxygen content could be determined accurately, the experiments were performed in such a way that the animals but exceptionally consumed 50 per cent of the oxygen content of the bottles, generally 25 per cent or still less. Frequently, the animals themselves secured sufficient circulation in the bottles; if this was not the case, the bottles were at intervals rotated cautiously. The analyses of water samples drawn directly from the large container and from the controls always showed satisfactory agreement.

2.15. Weighings.

As mentioned previously, it was feasible to weigh macro-organisms, only. For the sake of comparison with earlier investigations from this field, the animals had to be weighed in a fresh state, *i. e.* slightly dried on filter paper. In the case of molluscs, the tissue was dried and weighed immediately after removing it from the shells. Obviously, this method of determining the weight of the respiring tissue is encumbered with great uncontrollable errors, because a greater or smaller quantity of water remains on the weighed animal or tissue. This source of error, however, is common for these measurements and similar ones performed by others.

2.16. N-Determinations of the Order of γ .

After the conclusion of a respiration experiment in a μ l diver the animals were removed from the diver for the N-determination *ad modum* BRÜEL, HOLTER, LINDERSTRØM-LANG and ROZITS (15). The seals in the diver neck were removed with the aid of filter paper; the neck was rinsed with water and, subsequently, the total content of the diver was transferred into a salt cellar by means of a pipette filled with water, introduced into the bottom of the diver and blown out cautiously. The size (length) of the animals was then measured

under the microscope and the animals were transferred into special ampullae along with as little water as possible (usually c. $1\ \mu\text{l}$). The destruction of the organic material was performed in the ampullae. Pipetting was carried out, using a fixed stand for the pipette and a movable rack and pinion clamp for the ampullae, analogously to the arrangement used for the charging of the divers as described by HOLTER (41). Immediately after the transfer of the animals, $4\ \mu\text{l}$ of the digestion mixture ("blue mixture") were pipetted into the ampullae which were suitably labelled. All ampullae resulting from one day's work were sealed into a large ampulla (test tube) and stored until analyses could be performed at the Carlsberg Laboratory. The ampullae should not be touched with the fingers but, apart from this it has not always been practicable under the primitive conditions prevailing in Helsingør to satisfy the severe claims of utmost cleanness observed at the laboratory in Copenhagen. This point will be discussed later.

The analyses proper were performed at the Carlsberg Laboratory according to the following method. The samples are dried over P_2O_5 in a dessiccator. During drying, the organic material is carbonized by the sulfuric acid of the digestion mixture. Subsequently, $1\ \mu\text{l}$ H_2SO_4 conc. is added and the samples are boiled on a sulfuric acid bath ($1\ \text{ml}$ $\text{H}_2\text{SO}_4 + 0.4\ \text{g}$ K_2SO_4) for 6 hours at 295°C . The ampullae are open so that water and SO_2 evaporate¹. After cooling, the content of the ampullae is transferred to the bottom of a dwarf test tube coated on the inside with paraffine. So much water is used for the washing that the dwarf test tube contains c. $21\ \mu\text{l}$ of liquid. Finally, $9\ \mu\text{l}$ $18\ \text{n}$ NaOH are added. The neck of the dwarf test tube is closed immediately with a water seal ($45\ \mu\text{l}$), and $7\ \mu\text{l}$ of a $0.0145\ \text{n}$ H_2SO_4 are added to the seal. The tube is covered with a rubber cap and is placed in a water bath at 40°C for 1.5 hours. However, only the lower part of the vessel containing the bottom drop is immersed into the water bath. The temperature difference thus established between acid seal and bottom drop markedly promotes the distillation of NH_3 in the direction of the receiver acid. The not neutralized fraction of the receiver acid, however, is not titrated directly with

¹ Some of the 1943 experiments were carried out using destruction in closed ampullae as described in (15). This procedure was abandoned later and it has contributed to the fact that almost all 1943 experiments had to be discarded. Only the experiments on *Mytilus* of a length of 1–2 mm were used. They are uncertain.

base, because the latter will corrode the glass of the micro-burette. Prior to titration, an excess of alkaline buffer is added to the receiver acid which is then titrated back with $n/100$ HCl to the colour shift of brom cresol green (p_H 4.6). The base added ($20 \mu l$) is a phosphate solution ($m/150$ Na_2HPO_4) of such a pH that the bound NH_3 is not driven out. The indicator is dissolved to the same concentration in the receiver acid and in the titration acid. The N-content of the sample in γ = titration value of the analysis in μl minus that of the control times 0.140.

When making the analyses, however, it was found necessary to introduce certain modifications to be described below. Generally, it holds for all chemical analyses that the quantity of the substance present in a sample should be roughly known in advance. However, it was the aim of the present study to determine the respiration per N of the greatest possible number of different animals and especially such animals the size of which should vary within wide limits. During a three months stay in 1944 at Helsingør, moreover, the respiration experiments on micro-animals were accumulated, although it was impossible simultaneously to gain information concerning the N-content of the respective animals, because the N-analyses had to be postponed.

The experiments performed in Helsingør could not be repeated without rearranging the experimental setup in the following summer, which was not considered appropriate. It, therefore, became very important not to loose samples during the procedure. Thus, on the basis of orientating experiments from 1943, preliminary and approximate relations between the respiration and the N-content of the animals were established. From these relations the approximate N-content of the 1944 samples could be estimated on the basis of the respiration measured. The estimate was always made high because a successful analysis is secured only if the sample does not contain more N than expected, while it is of minor importance whether it contains considerably less.

2.161. *Modification of the Receiver Acid and the Titration.*

The quantity of receiver acid prescribed in the Carlsberg method (15) is completely neutralized by 1.4γ N (as NH_3) and, therefore, the upper limit for the applicability of the method is stated by the authors to be somewhat lower, *viz.* c. 1γ N. In many cases, it was

found necessary to increase the quantity of receiver acid, because the possible N-content of the sample was estimated considerably higher than 1%. Consequently, the samples were arranged in three groups, where 1, 3, or 6 times the ordinary quantity of receiver acid and of phosphate were used.

The analyses performed with once the quantity of receiver acid were thus exactly as described in (15). In the 3 times group, the same volumes of receiver acid and of phosphate were used as in the first group, however both in the threefold concentration; also the normality of the titration acid was 3 times that usually applied. Finally, in the 6 times group, the same receiver acid and phosphate as in the 3 times group were used, however taking 2 volumes of each. Titration was carried out with the same acid as used in the 3 times group. In all experiments, the concentration of the indicator was the same as stated in (15).

Thus, in most cases, the amount of receiver acid was sufficient. In very few cases only, the samples contained NH_3 in excess of the acid. After distillation of these samples we may assume the still free NH_3 to be evenly distributed over the sample and the neutralized receiver acid; moreover, a small fraction of the free NH_3 might have been lost to the air. However, in view of the enormous solubility of NH_3 in water, the loss is presumably very small. Therefore, in a few cases, an attempt was made to save the samples; after the titration had revealed that all receiver acid was consumed, an additional volume of acid was added and distillation was repeated for another 1.5 hours. After a second titration, the N-content of the sample was calculated on the assumption that the loss of N to the air was negligible. One single control experiment with this procedure, included in Table 1, seems to indicate that the method was applicable.

Table 1 shows a number of N-analyses performed on a varying number of *Nassa* embryos taken from the same cocoon, either A or B (both cocoons presumably originating from the same mother-animal). It seems to be unessential whether the analyses are performed in the ordinary way (*i. e.* as outlined in (15)) or with 3 times or 6 times receiver acid or with 3 times plus extra receiver acid and repeated titration.

Except for the last of the above mentioned modifications, the procedures chosen were presumably unassailable. Receiver acid, phosphate, and titration acid were made stronger than normal exclusively for the sake of security. When the samples nevertheless

Table 1.
Nassa reticulata.

Cocoon	Number of embryos	N/embryo	Method
A	6	0.133	ordinary
	18	0.126	6 times
	36	0.134	3 times + 3 times ¹
B	6	0.142	3 times
	15	0.132	3 —
	18	0.133	3 —
	21	0.125	3 —

¹ First titration 13.54 μ l titration acid.
Second titration 2.28 μ l titration acid.

proved not to contain more N than would have been determinable with the ordinary quantity of acid in the receiver, this only means that the increased security of the analyses has been paid with a slightly reduced accuracy of the final titration. However, this is less essential, because the accuracy of the N-analyses obtained was sufficient, as will be discussed later.

2.162. Destruction.

During destruction of organic material H_2SO_4 is reduced and evaporates as SO_2 . But the amount of H_2SO_4 is high enough to destroy even quantities of N $> 1\%$, as often present in the samples. In very few cases only, the N-content of the samples was so high that one or a few extra volumes of H_2SO_4 had to be added. In all cases, however, it was ascertained that destruction had been complete by checking after its termination whether there was acid in excess; moreover, it was ascertained that the samples remained colourless during several hours of boiling with excess H_2SO_4 . It is unessential that the quantity of free H_2SO_4 varies during destruction, since an acid reaction is always prevailing, because the destruction comes to a standstill when the p_H increases, *i. e.* when almost (but only almost) all free H_2SO_4 is consumed. (It should be kept in mind that the ampullae are not heated directly, but on a sulfuric acid bath at 295°C).

Destruction of animals weighing milligrams.

It may be emphasized again that it was the aim of the present investigation to study the respiration/N of animals differing as much

as possible in size. Therefore, it was often necessary to examine the respiration of single animals of medium size in especially large divers. It was to be expected that the N-determinations of these samples would encounter difficulties. In Helsingør, however, the animals were only placed in ampullae, just as the other N-samples, and the analytical difficulties were postponed to a later date. At the Carlsberg Laboratory, these medium large animals were destroyed with $19\ \mu\text{l}$ H_2SO_4 conc. in larger ampullae, as applied previously by MILTON LEVY (69), and using $78\ \mu\text{l}$ blue mixture. The ampullae were not heated directly over the flame, but in the holes of a copper block regulated to 295°C . After the destruction, $476\ \mu\text{l}$ H_2O were added, the sample was mixed carefully and, finally, $20.75\ \mu\text{l}$ were removed for analysis performed according to (15).

In this procedure, it was necessary to break the small ampullae in which the animals had incipiently been stored. The lowest part of the ampullae (4–5 mm long) containing the animal was placed in a large ampulla where destruction took place. Sulfuric acid was pipetted into the small one. Although the arrangement with two ampullae, one inside the other, undoubtedly reduced contact between the acid (which boiled out of the small ampulla rather rapidly) and the tissue to be destroyed, destruction proceeded satisfactorily, because sulfuric acid was transported as vapour everywhere in the lower parts of the large ampulla. When calculating the dilution of the acid digest, the whole quantity of H_2SO_4 added is assumed still to be present. This is not quite correct, because some H_2SO_4 has been lost as SO_2 ; on the other hand, the error is undoubtedly negligible, because the loss of sulfuric acid was undetectable. If all H_2SO_4 had evaporated the total error would be 4 per cent.

2.163. *Control Experiments and Accuracy Estimation.*

In each series of N-analyses blanks are included, *i. e.* ampullae containing blue mixture, but no animals. The blanks are submitted to the same procedure as are the N-analyses.

In (15) the authors claim that the work must be carried out in a dust-free and as far as possible NH_3 -free room, and they emphasize the necessity of protecting the pipette for the blue mixture and the sulfuric acid against NH_3 from the air. The significance of the last mentioned point was not yet clear when the work at Helsingør was going on, and the two first mentioned claims could not be fulfilled,

because in Helsingør the experiments were carried out in a dark, small, poorly ventilated cellar room. Next to the laboratory was the heating cellar, this being a possible source of NH_3 -containing air. When people in the upper apartment moved vivaciously around, the whitewash might often drop as dust from the ceiling. These circumstances, and also the extra manipulations resulting from storing the samples for 3–4 months before final analysis, may be the cause of the rather high scattering of the results from the series of blanks from Helsingør as compared with the data given in (15).

Table 2 shows the results of a number of series of blanks from Helsingør as compared with one series from Copenhagen. The blanks contain 4 μl blue mixture, but to the one denoted with *) also 0.5 μl sea water was added, this being the quantity of water generally pipetted into the ampullae together with the animals. However, sea water seems not to contain detectable quantities of N, as was to be expected. Moreover, in Parnas-Wagner determinations on 1–2 ml sea water from Helsingør, the author was unable to trace N with certainty. KROGH and KEYS (62) found only c. $2.6 \cdot 10^{-4}$ γ N per μl filtered, oceanic sea water. Of the 6 control series from Helsingør the first one ($^{19}/_7$ A) was determined with ordinary receiver acid, phosphate, and titration acid, as prescribed in (15). The following 4 were determined according to the 3 times, and the last one according to the 6 times principle. Within each series the scattering of the burette readings is not quite small. The standard deviation ΔK is calculated for each series and the corresponding nitrogen value is given below. The single series are so small because, incipiently, they were not intended for this purpose. However, the experiments show the standard deviation to be independent of whether the samples are determined as 1 x, 3 x, or 6 x. Evidently, the main reason for the scattering of the blanks is not to be found in the destruction or titration, but rather in the earlier stage of the procedure which took place in the Helsingør laboratory. This contention is supported by the fact that, accordingly, we find the results of the control series (Table 2) carried through at the Carlsberg Laboratory to scatter much less. The low N-content of the larval shells could only be determined in the Carlsberg Laboratory (Table 2). The conditions for work in Helsingør have thus reduced the accuracy of the analyses to one half or one fourth of what would have been reached at the Carlsberg Laboratory.

Thus, all samples from Helsingør, both the blanks and the

Table 2.

Helsingør						Copenhagen	
$19/7$ A (ordinary)	$24/7$ 3 x	$26/7$ 3 x	$9/8$ 3 x	$22/8$ 3 x	$27/7$ 6 x	ordinary	
2.74	2.18	2.30	2.40	2.22	4.48	2.72	
3.06	2.20	2.22	2.44	2.26	4.46	2.72	
2.92	2.22	2.24	2.32	2.26	4.50	2.66	
2.84*)	2.20	2.24	2.36	2.22		2.74	
M: 2.89	M: 2.20	M: 2.25	M: 2.38	M: 2.24	M: 4.48	M: 2.71	
$\Delta K = \pm 0.135$	$\Delta K = \pm 0.018$	$\Delta K = \pm 0.035$	$\Delta K = \pm 0.048$	$\Delta K = \pm 0.024$	$\Delta K = \pm 0.0163$	$\Delta K = \pm 0.035$	
$\sim \pm 0.019\gamma N$	$\sim \pm 0.008\gamma N$	$\sim \pm 0.015\gamma N$	$\sim \pm 0.020\gamma N$	$\sim \pm 0.010\gamma N$	$\sim \pm 0.014\gamma N$	$\sim \pm 0.005\gamma N$	
$\sigma = \pm 0.027\gamma N$	$\sigma = \pm 0.011\gamma N$	$\sigma = \pm 0.021\gamma N$	$\sigma = \pm 0.028\gamma N$	$\sigma = \pm 0.014\gamma N$	$\sigma = \pm 0.020\gamma N$	$\sigma = \pm 0.007\gamma N$	
Shells		γN					
6 Mytilus		0.077	± 0.007				
5 —		0.063	± 0.007				
4 1/2 —		0.080	± 0.007				
2 Nassa		0.058	± 0.007				

analyses, are encumbered with an error as stated for the blanks in Table 2. Since the true titration values for N in the analyses appear as the difference between the titration values of the analyses and the average titration value of the blanks, the standard deviation of the individual analysis is described by the figure σ , appearing in Table 2 as the standard deviation for the single blank times $\sqrt{2}$.

The value for σ varies from series to series (cf. Table 2), but we cannot decide whether this variation is significant. This is not improbable, however, and we may reasonably assume the conditions for N-analyses in the Helsingør laboratory not to be equally unsuited every day; they may, for example, have varied with the degree of ventilation in the cellar. For this reason, and in view of the deficiency of the whole material, we should reasonably assume a rather high value for σ , for example $\pm 0.03 \gamma$ N.

We therefore adopt the following standard deviations for the single analyses started in Helsingør.

Table 3.

< N <	$\pm \%$ σ $> \pm \%$	% of all the analyses
0.3 γ	10	5.1
0.3 γ 0.5 -	6	6.9
0.5 - 1.0 -	3	19.0
1.0 - 1.5 -	2	20.1
1.5 - 3.0 -	1	24.0
3 - 5 -	} $\sigma > \pm 1-2 \%$ }	5.1
5 - 10 -		8.1
10 - 20 -		2.3
20 - 110 -		9.2

The material consists of about 200 analyses, the blanks not included. The table also shows the distribution of the analyses with regard to the N-content. Only 5 % of the analyses show an N-content $< 0.3 \gamma$ and only for these we must reckon with a standard deviation $\geq \pm 10 \%$. All other analyses are regarded as sufficiently accurate and they are published with the single remark that only 7 % of them are encumbered with an error $> \pm 6 \%$, the majority of the analyses being considerably more accurate.

Table 3 does not comprise the experiments from Copenhagen, discussed earlier in Table 2, dealing with the N-content of the shells of *Mytilus*- and *Nassa* larvae. The animals were subjected to auto-

lysis for two months in sea water so that only the shells remained. Perhaps, however, the conchioline (a scleroproteid) is still present in the shells which appear yellow, just as they do on the living larva. The conchioline covers the surface of the shells of the adults and, at least in nature, it stands exposure to sea water for years. The N-content of the larval shells was low, but it could be determined with reasonable accuracy, as appears from Table 2 (cf. Fig. 27).

A special control experiment for the N-analyses of the Helsingør material is shown in Table 1. The embryos were removed from two cocoons of the prosobranch gastropod *Nassa reticulata*. On these embryos a series of determinations were made, using increasing numbers of larvae. The results do not seem to indicate differences between the embryos from the one or the other cocoon. The reason for this presumably is that the cocoons are produced one immediately after the other by the same animal. Considering all experimental results represented in Table 1, we arrive at a somewhat higher standard deviation ($\sigma = \pm 0.056 \gamma \text{ N}$) than found on the basis of the control experiments of Table 2 alone ($\sigma = \pm 0.03 \gamma \text{ N}$). This might seem strange, but actually we know that the *Nassa* embryos employed were not equally large. This, of course, must contribute to a scattering of the results obtained from analyses in which limited numbers of animals are used. Since, however, the variation in size of the embryos remains unknown, it is impossible to estimate to which degree this factor contributes to the scattering of the results. Finally, it must be mentioned that the embryos were embedded in mucous secretion of the cocoon, a fact not sufficiently taken into account by the author when the experiments were carried out. The embryos moving out of the cocoon into a large volume of water were, therefore, perhaps not sufficiently washed before the samples were taken. None of the other animals investigated was in a cocoon containing secretion. For these reasons, the error estimation given in Table 2 should form the basis of the estimation of the error of the whole material. Table 1 mainly serves to demonstrate that samples can be saved—as discussed on p. 36—by addition of more receiver acid and phosphate; the table is, therefore, published in this connection.

Unfortunately, Table 1 contains the only series of control experiments carried out in Helsingør on a series of samples, the relative N-content of which is known. As already mentioned previously, destruction and titration of the samples were always carried

out at the Carlsberg Laboratory, where due regard was paid to all precautions prescribed in (15). For this reason, the control experiments performed by the Carlsberg workers are applicable also to the present destructions and titrations. This, however, does not hold for samples containing more than 1γ N; for these determinations, on the other hand (up to 6γ N), Table 1 is a quite satisfactory control, the standard deviations being at the most $\pm 5 \%$. As regards the samples which, because of their high N-content, were digested with $19 \mu\text{l}$ H_2SO_4 , they scarcely need any special control. The reader may be referred to LEVY's (69) paper and to the remarks made previously. The analyses of samples drawn from the same diluted acid digest always agreed satisfactorily.

2.17. N-Determinations of the Order of mg.

It appears from the preceding section that, after introducing minor modifications, the Carlsberg micro-method can be applied to the determination of N in samples containing $1/10$ -c. 100γ N. Samples the N-content of which was assumed to be between $1/10$ -c. 1.5 mg N were analyzed according to PARNAS and WAGNER (88). Animals containing still more N were initially hydrolyzed by heating with 50% sulfuric acid. The hydrolysate was subsequently diluted in a measuring flask and a suitable fraction was removed for destruction, distillation, and titration according to (88). The analyses of samples containing very little N (0.1 mg) can be considered accurate to $< \pm 10 \%$; the majority of the determinations, however, is encumbered with a much smaller error. Since the analyses were performed exactly as prescribed by the respective authors, a further discussion of the procedure seems superfluous.

2.18. Determination of the Respiration/N.

From the work carried out at the Carlsberg Laboratory we have gathered knowledge concerning the errors involved in respiration measurements with the Cartesian diver of the μl type. The preceding discussion of the material accumulated in Helsingør offers an impression of the errors involved in the N-analyses. Unfortunately, no experiments were performed in Helsingør with the aim directly to determine the error of the respiration/N measurements, because

among the numerous objects of the investigations none was found suited for such a test. The main difficulty was not to find sufficiently uniform animals, but to find animals which in parallel experiments would react with the same activity and, hence, with the same metabolic rate to the experimental conditions.

However, the author does not believe to have overestimated the methods involved when assuming in general to operate with an error of $\pm 10\%$ or less for both methods used. Only very rarely therefore, the error of the single respiration/N values will amount to $\pm 20\%$. In the present paper, generally no conclusion will be drawn beyond this limit of accuracy. This decision is very easy to make, since from the following section it will appear that the mode of reaction of the animals to the experimental conditions is so many-fold (the "biological scattering" of the results therefore so high) that this fact alone will prevent us from placing too much weight upon small differences found for the respiration/N.

As regards the accuracy of the values found for the respiration/N it may be emphasized that the scattering of the results depends on the animal species studied. It will, thus, be natural to assume that the accuracy of the analyses is even higher than indicated by the results found on such animals (*Asterias* larvae, Fig. 21, and *Vermes* larvae, Fig. 22), where the scattering is smallest. These experimental results seem to indicate that the experimental error is considerably below $\pm 20\%$, even in experiments on animals other than those just mentioned.

2.2. *Biological Factors in the Metabolism Measurements.* *Planning of Experiments.*

As soon as the experimental technique works satisfactorily, purely biological difficulties again become dominant. These difficulties are obvious. All investigators who have measured O_2 -consumption of marine invertebrates will know that the figures obtained on a number of animals of the same species, of the same size, and from the same locality almost always show an unpleasantly high scattering—frequently amounting to 200–300%. This is especially disappointing if we realize that the metabolic rate of man under standard conditions can be reproduced from one individual to the other with an accuracy of about 10%. Even during work of a well-

defined intensity, the metabolic rate of different human subjects is well comparable.

In the animals studied here, the respiratory metabolism was usually found to be very labile, presumably owing to the fact that usually we are unable reasonably to adjust the exterior and interior factors which influence the metabolism. Some of these factors will be discussed in the following.

2.21. Temperature. The temperature is the only of the many factors acting upon the metabolism of animals, the effect of which is rather generally and accurately known to us. The majority of the cold-blooded animals seems to follow a metabolism-temperature curve of the same type as KROGH's (59) standard curve. But, moreover, the temperature has an indirect effect upon the respiratory metabolism. If a cold-blooded animal is transferred to a lower temperature, the metabolic rate will be found to decrease immediately in accordance with KROGH's curve. However, after having been kept at the lower temperature for some time (20–30 days), the animal has often actively regulated its metabolism to a somewhat higher value. Reversely, the same animals are able to regulate their metabolism downward after they are kept at a higher temperature for some time (DOUDOROFF (25) on the fish *Girella nigricans*).

2.22. Activity. The activity often considerably influences the metabolic rate of animals. Thus, in comparing the metabolism of different animals, the investigators have usually attempted to study animals which are in the same state of activity. However, the only well-defined state of activity common for different species is the activity = 0. The concept standard- or basal metabolism originates from human physiology, since the metabolism of humans studied under standard conditions (defined as absolute muscular rest at a convenient temperature and fasting) is constant and well reproduceable even from individual to individual. The metabolism of cold-blooded animals never seems to assume such a well-defined basal level. At rest and during fasting the metabolism often decreases continuously. In comparative physiology, the concept "standard metabolism", therefore, is seldom of any meaning, especially if widely different species are compared, common "standard conditions" for such animals hardly being definable.

In the present experiments, the claim of "standard conditions" has therefore been abandoned and, instead, it has been attempted to compare the animals at as far as possible "normal activity". Hereby, we mean a "visible activity" approximately corresponding to the activity regarded by the investigator as being characteristic of the respective animal in nature at 16° C. The draw-backs of this subjective definition of "normal activity" are quite obvious but, unfortunately, they will probably be common for all comparative-physiological investigations of the metabolism of animals. The important consequence is involved that the figures obtained (encumbered with the below discussed uncertainty) must be considered valid also in nature. The problems concerning metabolism and activity are discussed again under 3.11.

2.23. Food ingestion. In all animals, the ingestion of food causes the metabolism to increase, partly because the work of the intestine and, thereby, the metabolism of this organ is increased, partly, because of the specific-dynamic (*i. e.* accelerating) effect of the food. While the metabolism of some animals is multiplied (snakes, c. 3—8 times, BENEDICT (3)) after feeding following a period of starvation, other animals (*Planaria*, ALLEN (1)) show a more moderate, however considerable increase in O_2 -consumption after feeding. For the species studied in the present work, we may assume that all of them feed rather continuously in nature and, therefore, they contained food in the intestine when they were collected. The respiration of the animals was not measured before about 2 hours later and it seems difficult to decide to which extent the increase in metabolism due to feeding had disappeared at the start of the respiration experiment. The respiration sometimes showed a decreasing tendency in the course of the experiment.

2.24. Light. Light affects the metabolism of many animals; thus, for example, the metabolic rate of *Calanus finmarchicus* is highly dependent on the light (MARSHALL and NICHOLLS (80)). The British investigators correlate the light sensitivity of the metabolism with the diurnal migrations of the copepods up and down the plankton. Such light-sensitivity of the metabolism of the present animals can certainly not be excluded.

2.25. Rhythms. The metabolism of numerous animals shows a diurnal rhythm. This is well known in the case of higher animals in which the metabolism is low during that part of the day when the animals are normally asleep, and higher when the animals are normally awake and active.

Even on lower animals such rhythms have frequently been observed (LANG on *Lumbricidae* (68), MICHAEL on *Tenebrio* larvae (82)). Of special interest are the rhythms observed by GOMPEL (30) on a number of animals from the littoral zone. The metabolism is considerably higher during high-tide than during low-tide, and the metabolic rhythm is maintained even after the animal is no longer subjected to the tide. Numerous other rhythms might affect the metabolism of the animals (measured at a given temperature), such as the moon cycle, the seasonal cycle, and the sexual rhythms of the animals. Frequently, the animals show an increased metabolic rate during the period of reproduction (cf. THORSON on bivalves (108), and others).

For the animals studied in the present work many such rhythms must be considered possible. There is no reason to assume that the different species were in the same phase of their possible rhythms when the metabolism was measured. The rhythms, therefore, may contribute to the scattering of the results obtained, especially when different species are compared.

2.26. The geographical latitude and the season of the year also play a part in the metabolism of the animals. In both cases, the reason seems to be that the temperature at which the animals live calls forth regulatory mechanisms (SPÄRCK (103), FOX and co-workers (27, 28, 117), IRVING and associates (26, 38)), tending to increase the metabolism at low temperature and to decrease it at high temperature. This fact should be kept in mind when

comparing cold-water and warm-water forms taken from waters of different temperatures.

2.27. Within physiological limits, the oxygen pressure has scarcely any significance for the micro-fauna of the plankton, however, possibly for the bottom fauna. A few experiments are discussed under 3.12.

2.28. The study of the significance of the body size for the metabolism of animals is the present author's main problem which will be treated in greater detail under 4 and 5, where also the influence of other factors will be discussed.

2.29. The position of the animals in the zoological system is of significance for their metabolism, as it will appear from 4.

The influence of various other factors, such as the sexual state, age, rate of growth, possibly the living habit (sedentary or planktonic, rapacious animals or herbivorous animals) will not be mentioned or only be touched upon.

A discussion of the above mentioned factors (2.21—2.29) was found necessary, both because many authors do not realize the principal difficulties involved in comparative respiration measurements, and also in order to avoid misinterpretations of the author's own measurements, the uncertainty of which should be stressed.

In the present treatise, all figures will be published in such a way that the scattering of all single determinations obtained on the same species becomes apparent; it should be emphasized that each average figure may possibly be wrong to an unknown extent, because each species undoubtedly reacts characteristically to the given experimental conditions which, of course, always deviate from those prevailing in nature. This possible species-characteristic reaction of the metabolism towards the experimental conditions must be kept in mind, especially when comparing different species with one another, and also when comparing the larvae with the adult forms which morphologically and physiologically may often be only remotely related. The present author considers large and repeatedly observed differences between the metabolic rate of two species to be more indicative of similar differences, also in nature, than are correspondingly small ones, and he therefore prefers to draw conclusions only on the basis of large differences. Small differences will generally be stated without comment; they can frequently be met with in the tables and the figures of this paper.

According to the author's view, the biological difficulties are dominant as compared with the technical ones, and they invariably involve an uncertainty of the single determinations which will only be reduced in so far as we may succeed in finding a systematic valuation of the figures obtained for the metabolism of widely different animals.

3. EXPERIMENTAL PART

3.I. *Special Problems Concerning the Metabolism of the Micro-Fauna.*

3.II. Metabolism During Natural Activity Variations.

When performing a series of successive measurements of the oxygen consumption of one and the same individual, a satisfactory agreement of the results is found on some species while, on other species, a considerable variation is observed which—as far as it can be judged—varies in accordance with the visible activity of the animal.

3.III. *Echinodermata and Vermes.*

Among the plankton types of *Echinodermata* and *Vermes*, both the respiration and the visible activity seem most frequently to remain constant throughout the whole period of experiment. When the respiration changed markedly, some injury of the animal could generally be detected. Fig. 4 shows four experiments carried out with one worm in each and, moreover, one experiment with three *Asterias*. Long-lasting experiments on these animals were performed in rare cases, so that the constancy of the respiration could only be illustrated by few experiments. This, of course, was done because not even short-lasting experiments ever showed indications of a systematic decrease in the metabolic rate during the experimental period.

Obviously, the experiments only indicate the oxygen consumption of the small forms not to vary considerably with time. On larger animals, where respiratory movements and other respiratory mechanisms may be necessary in order to secure an adequate oxygen

supply, the oxygen consumption may easily fluctuate with the efficiency of these mechanisms (cf. LINDROTH (76)).

In the author's opinion, the relative constancy of the metabolic rate in *Echinodermata* and *Vermes* is mainly due to the fact that,

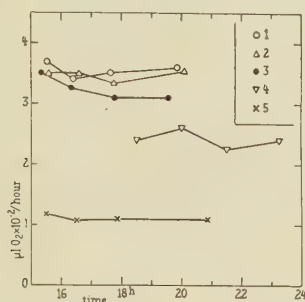


Fig. 4. Constancy of the respiration intensity of different lower animals.

1. 1 *Magelona* (length = 0.9 mm).
2. 1 *Polydora* (l. = 0.7 mm).
3. 1 *Polydora* (l. = 0.6 mm).
4. 1 *Phyllodoce* (l. = 0.75 mm).
5. 3 *Asterias* (respiration/larva) (l. = 0.6-0.8 mm). μ l divers.

usually, the ability of these animals to alter their activity is not very great. We here have to do with rather indolent animals with a weak muscular system and a rather low metabolic rate, the main part of which is "basal". The experiments are possibly indicative of the conditions prevailing in most lower, small, relatively passive invertebrate animals.

3.II2. *Mollusca*.

The mollusc veliger larvae exhibit a marked ability of varying their visible activity. Sometimes, they swim vividly, sometimes they retract the velum and settle on the bottom of the diver, resting more or less deeply. Even in the resting larvae, however, the cilia beat more or less, but after a long period of rest the activity within the shells can be very low, and this is true both for the velum, the foot, and the cilia on the gills. On the other hand, resting larvae may gradually resume their swimming activity. For a short time they extrude the velum half way out of the shells, move the cilia a little, but immediately stop again. This is repeated at still shorter

intervals; however, the animals do not swim. But finally, they depart, beautifully swimming.

The metabolic rate corresponding to different states of activity was measured in a series of experiments with single larvae in the $0.1 \mu\text{l}$ diver. Some of these experiments and a few performed with the μl diver are shown in the following.

Bivalves:

In Fig. 5 three simultaneous experiments on three lamelli-branch larvae are recorded. The two lowest curves represent the results with two single larvae of *Mya arenaria* and *Zirphaea crispata*. Both animals were at rest during the whole experimental period,

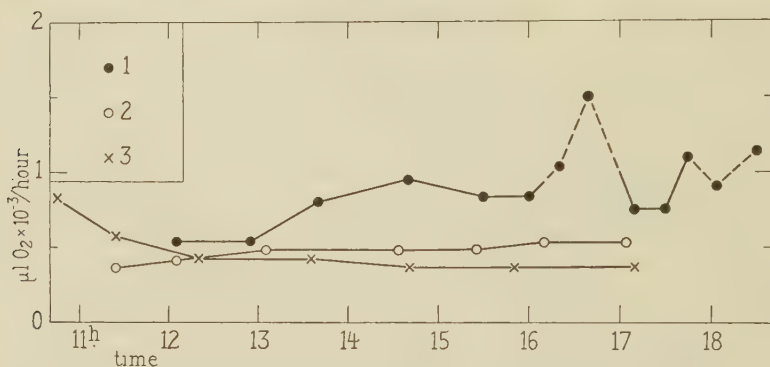


Fig. 5. Lamelli-branch veliger larvae from the plankton. The variation of the respiratory rate during long lasting experiments ($0.1 \mu\text{l}$ divers).

Periods of swimming dotted, periods of rest fat. In this and in the following figures (5-15 incl., and 17) the respiration values measured are plotted in the middle of the corresponding experimental period.

1. *Mytilus edulis* (length = 0.235 mm).
2. *Zirphaea crispata* (l. = 0.214 mm).
3. *Mya arenaria* (?) (l. = 0.170 mm).

although, when introduced into the diver, they were swimming; their metabolism remains rather constant for c. 6 hours.

The uppermost curve shows an experiment on a young *Mytilus* larva which was at rest from 12 o'clock to 16¹⁰. However, to judge from the curve representing the metabolism, the rest was not deep after 13 o'clock. Between 16¹⁰ and 17⁰⁰ the animal was almost incessantly swimming, and the metabolic level is increased. Between 17⁰⁰ and 17⁵² the animal did not swim and its metabolism is again

somewhat lower. From 17⁵² until the end of the experiment the animal swam anew, the metabolism, however, not being definitely higher than the highest resting values. On the whole, swimming is accompanied by a high metabolic level, but the curve seems to indicate the condition of so-called "rest" possibly also to include periods of activity other than swimming. Later experiments (cf. 3.143) might indicate this other activity to be ventilation of varying intensities by means of the gills. However, this experiment was performed at such an early stage of the present investigation that the

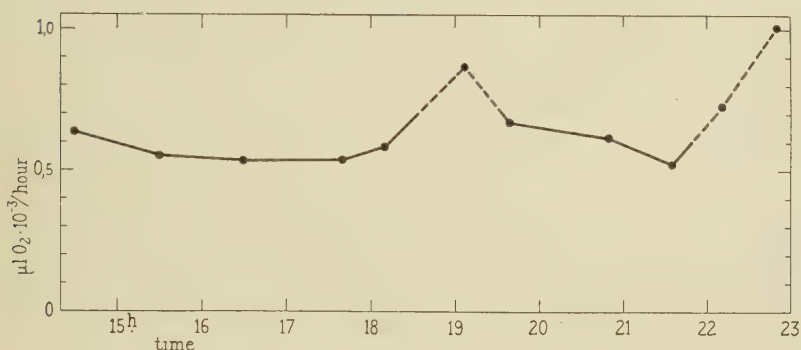


Fig. 6. Unidentified lamellibranch veliger from the plankton.

The metabolism increases as the animal starts creeping on the foot (dotted). Fat curve: periods of rest. 0.1 μ l diver. Length of animal 0.25 mm.

author's attention was not sufficiently directed towards closely observing the behaviour of the "resting" larva. In any case, however, the larva was not seen to move on the foot.

Fig. 15, Section 3.12, represents an experiment with a larva of *Mya arenaria*, this individual being extraordinarily liable to swim. Only during a short period (around 23 o'clock) it rested on the bottom of the diver and during that period the metabolic rate was about half that during swimming. The subsequent decrease in metabolism (dotted line) must be ascribed to a simultaneous decrease in oxygen pressure, as it will be discussed in 3.12.

The reader should also be referred to the Sections 3.12 and 3.143 which include experiments showing how the metabolic rate in lamellibranch larvae may vary.

Not only during swimming (and during ventilation by means of the gills, as described in 3.143), but also when the larva crawls on the foot, the oxygen consumption increases. An experiment with

an unidentified bivalve larva illustrates this observation (Fig. 6). During long periods, the larva lies on the bottom of the diver, during 2 periods it moves vividly on the foot, and the metabolic rate therefore increases. No information can be given concerning the possible ventilation activity, neither at "rest" nor during crawling.

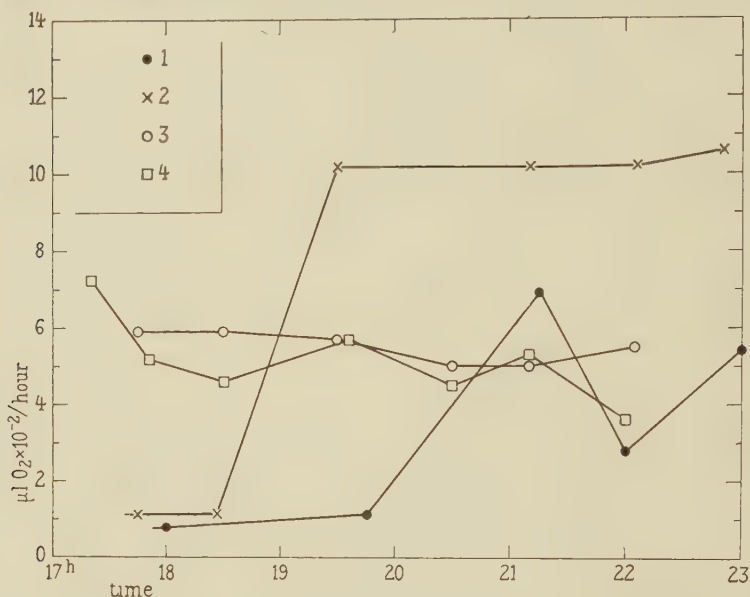


Fig. 7. 4 young individuals of *Mytilus* from the bottom.

The metabolism may vary enormously with time (μ l divers).

1: length = 1.25 mm.

2: l. = 1.48 mm.

3: l. = 1.14 mm.

4: l. = 1.31 mm.

Finally, Fig. 7 records four experiments in μ l divers with 4 young sedimented *Mytilus*, one in each diver. Two of these experiments indicate relatively constant oxygen consumptions, while the other two show considerable variations with time of the metabolic rate. During the periods in the beginning of the experiments, when the oxygen consumption was lowest, the shells were completely closed, or nearly so. The experiments may be explained by assuming that the chalk shells of these small animals are so slightly permeable to O_2 that their closure involves almost completely anaerobic conditions. Thus, the author regards the closing of the shells as the

primary cause of the reduced oxygen consumption. The decreased oxygen supply may secondarily give rise to a reduced activity in the tissues, for example of the cilia of the gills. This interpretation offers the simplest explanation of the phenomenon described earlier, *viz.* that in small veliger larvae a long period of shell closure implies a gradual reduction of the visible activity within the transparent chalk shells (lack of oxygen and accumulation of metabolic products). This also agrees with the fact that the metabolic rate of closed, resting larvae can be very low. Finally, Fig. 7 demonstrates how carefully respiration experiments on lamellibranchs should be evaluated, even when numerous animals are used in each experiment.

A few experiments showed that the oxygen consumption of closed adult bivalves is practically zero. This is not surprising and has been described repeatedly in the literature (cf. v. BUDDENBROCK's book (19)).

Gastropods.

Fig. 8 shows an experiment in the μ l diver with an unidentified gastropod veliger larva having an especially large bilobular velum.

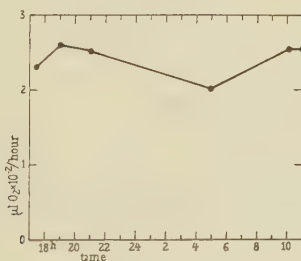


Fig. 8. Metabolism of unidentified prosobranch veliger larva from the plankton.

This animal has only been observed swimming. Observations, however, were interrupted during the night.

Largest shell diameter = 0.406 mm (μ l diver).

Each time the animal was observed, it went on swimming; however, it was not observed between 24 o'clock and 9³⁰; possibly, the animal has been at rest during part of that period. In spite of the fact that the animal appeared fatigued at the end of the experiment (the velum was extruded, cilia moved, but apparently more slowly than before, and the animal seemed unable to rise for swimming) the metabolic rate was rather constant throughout the whole period of swimming.

Fig. 9 represents two experiments in $0.1 \mu\text{l}$ divers on single *Littorina* veliger larvae. Swimming is marked by fat dots. In experiment A the metabolic rate is somewhat increased during swimming, but neither during swimming nor during rest the metabolic rate is constant; there are gradual transitions from highest and lowest resting metabolism to lowest and highest swimming

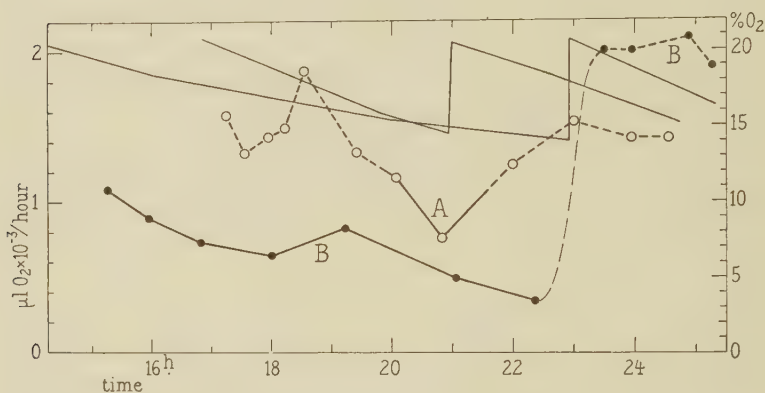


Fig. 9. Metabolism of 2 *Littorina* larvae (A and B) from the plankton.

Periods of swimming marked with strokes. Periods of not-swimming indicated by fat lines. ($0.1 \mu\text{l}$ divers).

Ordinate to the left: respiration.

Ordinate to the right: average oxygen pressure in the diver (the not denoted fat lines above).

The oxygen pressure increases owing to recharging of the diver (cf. the text). A is recharged first. The thin strokes of B indicate that, on the transition from rest to swimming, the metabolic rate increases, however following an unknown curve.

Animal A: largest shell diameter = 0.238 mm .

Animal B: „ „ „ = 0.256 mm .

metabolism. The most abrupt change in respiratory rate altogether observed by the author when experimenting on veliger larvae is shown in experiment B. This larva was at rest in the diver for 7 hours and then, suddenly, it started swimming; thereby, the metabolism increased to c. 6 times the values found at rest. This enormous increase should not be ascribed to an especially high metabolism of the swimming animal, but rather to an especially low metabolism of the resting larva. During the long-lasting period of rest, the metabolism gradually decreased, and it seems natural to assume that this

decrease follows a decrease in the visible activity within the shells, as it was frequently observed outside the diver on veliger larvae which had rested for a longer time. However, this is purely speculative, since an accurate observation could not be performed while the animal in question was in the diver.

Owing to the animal's respiration, the oxygen pressure inside the diver decreases. After animal *A* at 21 o'clock had been at rest for more than an hour, it was decided to introduce fresh air into L_3 , applying a technique which will be described later (cf. 3.12). Hereby, the oxygen pressure in the diver is brought back to the level prevailing in the beginning of the experiment, *i. e.* almost to the oxygen pressure of the atmosphere. However, already when the diver was removed from the vessel, *i. e.* before fresh air was introduced into the diver, the animal resumed its swimming activity, presumably as a consequence of mechanical or thermal irritation.

The same occurred later with *B* but, nevertheless, the introduction of fresh air was carried through in both cases. Therefore, the upper curves represent the change of the average O_2 -percentage in the divers during the experiment.

The influence of the O_2 -pressure on respiration will be discussed later, and it will become clear that the O_2 -pressure, both in *A* and *B*, throughout the experiment was unambiguously above the critical value, below which the respiration of swimming animals decreases with decreasing oxygen pressure. At the present place, the introduction of fresh air into the divers *A* and *B* is thus mentioned only because this detail cannot be omitted in a general description of the experiments. The question whether the decrease in respiration observed during the long-lasting period of rest of animal *B* can be ascribed to a decrease in oxygen pressure may be answered by the following statement: At 22²⁰—when the respiration of animal *B* had reached a minimum—the oxygen pressure at the surface of this animal was calculated to be 12.5 % of an atmosphere (in air 21 %). For the calculation (formula 6 in (118)) it was assumed that oxygen diffuses both through PO_2 and through that part of *W* which separates the centre of the animal from L_2 . At 12.5 % oxygen pressure the oxygen supply of the animal should be absolutely sufficient (cf. 3.12), unless the shell must be regarded as an especially effective brake against the diffusion-in of oxygen. The latter view cannot be excluded, and the low respiratory intensity measured after a longer period of rest may thus be due to oxygen deficiency in the tissues.

However, in this case, the primary cause of this oxygen deficiency must be that the resting animal actively cuts itself off from the oxygen by closing the shell by means of the operculum situated on the foot. Thus, we again meet with the problem of the oxygen permeability of the shell, just as mentioned above in connection with the lamelli-

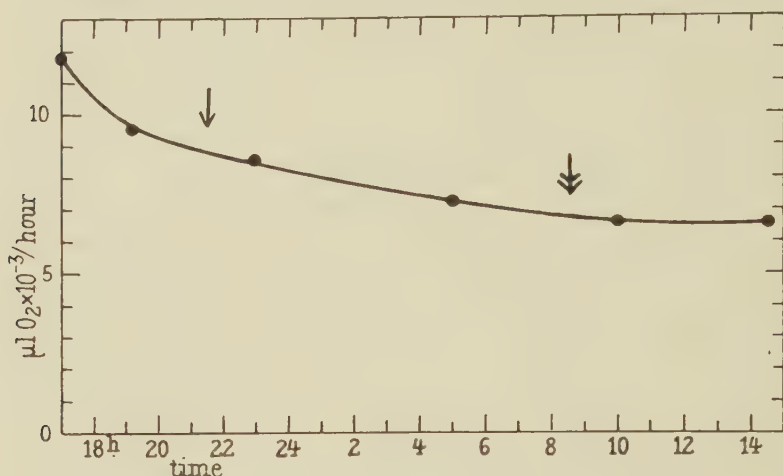


Fig. 10. Experiment with 3 *Nassa* larvae from the plankton.
Largest shell diameter = 0.40-0.43 mm. μl diver.

branches. The problem will be touched upon later (3.12) without, however, further approaching its solution.

An increase in respiratory rate at the onset of swimming or a decrease at the conclusion was repeatedly observed on veliger larvae. A few experiments performed on larvae of *Nassa reticulata* which are very liable to swim may be mentioned here. Fig. 10 shows an experiment on 3 *Nassa* larvae in a μl diver. In the beginning of the experiment all three larvae were swimming. At the moment indicated by the first arrow only one larva swims, and at arrow 2 all three animals are at rest. Fig. 11 illustrates an experiment with one large *Nassa* larva in a μl diver. At the start of the experiment the animal swam, but swimming stopped at the time indicated by the arrow. Figs. 10 and 11 show that the respiratory rate possibly decreases slightly at the transition from swimming to not-swimming, but the metabolism also decreases continuously after swimming has ceased.

In other experiments on gastropod larvae, the author repeatedly observed a sudden change of the metabolic rate at the transition

from swimming to not-swimming, this change amounting at the most to 50 % of the metabolic rate during swimming.

The metabolic rate during swimming of both lamellibranch and gastropod veliger larvae often varied very markedly, as it appears from Figs. 5 and 9. This may possibly be ascribed to varying

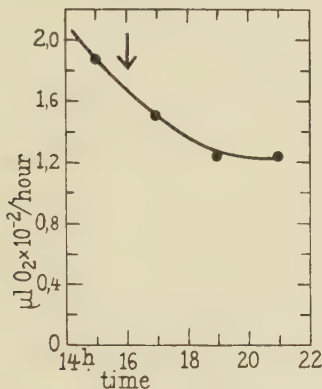


Fig. 11. One large *Nassa* larva from the plankton.

Largest shell diameter = 0.48 mm. μl diver. -

activity of the velum, but perhaps also to varying activity of other organs, for example the foot. Analogously, the considerable variations of the metabolic rate during rest may be explained by assuming a varying, however only partly visible activity of different organs. Consequently, we often find a rather smooth transition from the respiratory rate during swimming to that during not-swimming.

3.II3. Crustacea.

In some cases, the respiratory rate remained constant during the first 3–4 hours of the experiments; in other cases, it decreased already from the beginning of the experiment. Long-lasting experiments almost always show a decrease in respiratory activity, frequently coinciding with a decrease in the visible activity. Fig. 12 shows an experiment in a μl diver on a single individual of the copepod *Centropages hamatus*. Presumably, this experiment is rather characteristic of the small crustaceans of the plankton as a whole. As the experiment proceeds, the animal moves more rarely and more slowly, and the respiratory rate decreases slowly with decreasing

visible activity of the animal. At 23 o'clock the animal is at complete rest. Fresh air is introduced into the diver at the instant

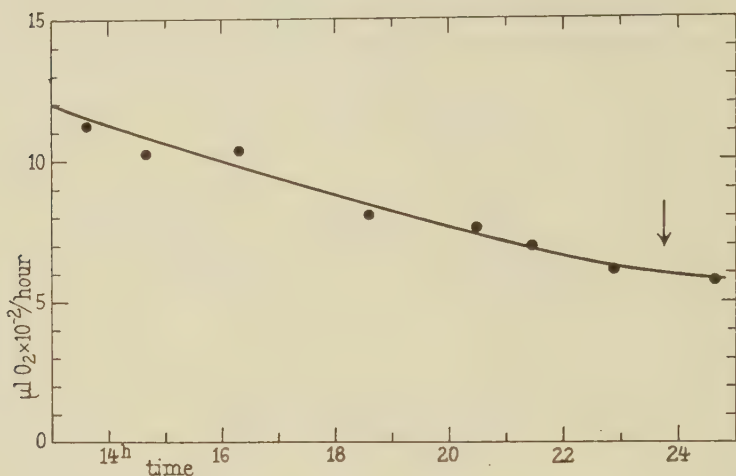


Fig. 12. Copepod from the plankton (*Centropages hamatus*).

Length of animal = 0,86 mm. μl diver.

indicated by the arrow. The metabolic rate remains at the low level. Thus, the animal was brought to rest neither by a beginning lack of oxygen nor by a factor accumulated in the air space of the diver.

3.12. The Influence of the Oxygen Pressure on the Metabolism and the Activity of Some Micro-Animals.

In the experiments with the 0.1 μl diver the respiration measured was frequently so intense that the oxygen pressure inside the diver decreased to a rather low value within a few hours. Thereby it became possible to study how the respiration depends on the oxygen pressure; simultaneously, the visible reaction of the animal to the decreasing oxygen pressure could be studied in the microscope.

At the moment when the charged diver is closed by the medium seals M_1 and M_2 the air space contains 20.9 % O_2 . At the start of the experiment, the diver floats at about $-20 \text{ cm H}_2\text{O}$. The average water vapour tension of the diver approximates $5 \text{ cm H}_2\text{O}$. The barometer was not read, but the pressure was assumed to be normal,

viz. 1030 cm H₂O. Thus, in the beginning, the oxygen pressure inside the diver is $\frac{(1030 - 5 - 20) \times 20.9}{100} = 210$ cm H₂O. A change of the equilibrium pressure of 10 cm thus corresponds rather accurately to a decrease in the oxygen pressure within the diver of 1% of an atmosphere. By extrapolation we can calculate the change of equilibrium pressure which would have been found had the change in equilibrium pressure been measured from the moment when M_1 and M_2 were placed in the diver and until the first reliable measure-

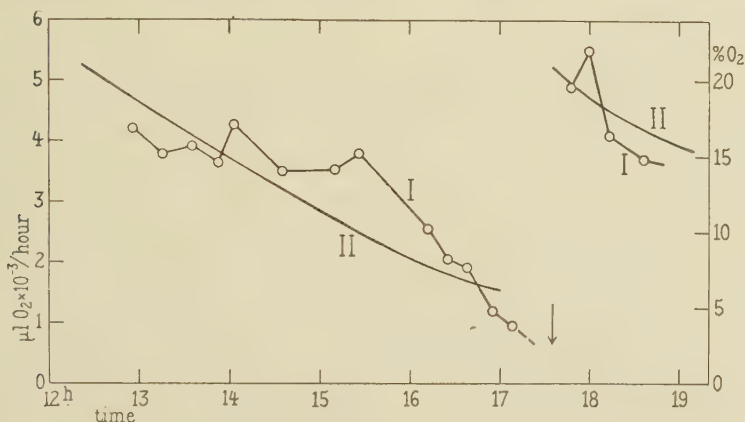


Fig. 13. Respiration and oxygen pressure. Trochophora from the plankton (*Pectinaria*).

Respiration (curve I and ordinate to the left), and average oxygen pressure in the diver (curve II and ordinate to the right). At the arrow, the air inside the diver is renewed. Length of the animal = 0.36 mm. 0.1 μl diver.

ment was obtained. As mentioned in (118), reliable measurements are only obtained when the "initial period" is over. From the change in equilibrium pressure of the diver, the average oxygen content of the diver at any moment of the experiment may thus be calculated with good approximation. However, as it appears from Fig. 2, p. 27, the total air space of the diver is divided in three spaces. The oxygen pressure inside the diver must have a minimum where oxygen is consumed, *i. e.* in the drop of water W . In L_2 the oxygen pressure is a little higher, and in L_3 it is still higher than this. Very soon, the difference in oxygen pressure between L_2 and L_3 will adjust itself to a constant value, depending on the magnitude of the respiration measured and on the conditions for diffusion through the

oil film PO_2 separating L_3 from L_2 . The oxygen pressure in L_1 is unknown to us, because it is difficult to estimate to which extent the solid paraffin in the complex $\text{P} + \text{PO}_1$ restricts diffusion of O_2 from this space to W .

It is of interest (cf. Figs. 13–15) to establish how the respiration intensity of the animal varies with the oxygen tension at the surface of the animal, *i. e.* with the oxygen tension in the water drop W . Directly, however, we can only compare the respiration measured (curve I) with the average oxygen pressure inside the diver as calculated from the change of equilibrium pressure and represented in curve II of the figures.

Table 4 shows the values of the average oxygen pressure within the diver at which the respiration began to decrease with decreasing oxygen pressure. The figures refer to the experiments illustrated by Figs. 13–15. Of course, the oxygen pressure in L_2 is lower than the average values given in the table, and the oxygen pressure in the metabolic centers in W is still lower. The formulae given in (118) render it possible quite approximately to calculate the difference between the oxygen pressures on both sides of the oil film PO_2 . If the oxygen pressure in L_3 is known at any moment of the experiment, we may, therefore, calculate the oxygen pressure prevailing simultaneously in L_2 . The average O_2 -pressure in L_3 is

Table 4.

Critical values of the O_2 -pressure in the divers
(exp. Figs. 13–15).

	Average O_2 -‰ in the diver ¹	Maximum O_2 -pressure in L_3
<i>Pectinaria</i> Fig. 13 $l = 0.36 \text{ mm}$	9 ‰	$9 - 2.6 = 6.4 \text{ ‰}$
<i>Phyllodoce</i> Fig. 14 $l = 0.39 \text{ mm}$	8 ‰	$8 - 1.8 = 6.2 \text{ ‰}$
<i>Mya arenaria</i> Fig. 15 $l = \text{c. } 0.2 \text{ mm}$	6 ‰	$6 - 0.9 = 5.1 \text{ ‰}$

¹ At the start of the experiment put to 20.9 ‰.

unknown, however that in $L_1 + L_2 + L_3$ is known, the latter being greater than the former after the experiment has been going on for a short time. Therefore, we shall arrive at an incorrect, however maximum value of the oxygen pressure in L_2 when assuming the pressure in L_2 to be the average O_2 -pressure in the diver minus the difference in O_2 -pressure on both sides of PO_2 . The value thus obtained is given in Table 4 as "max. O_2 -pressure in L_2 ". Evidently,

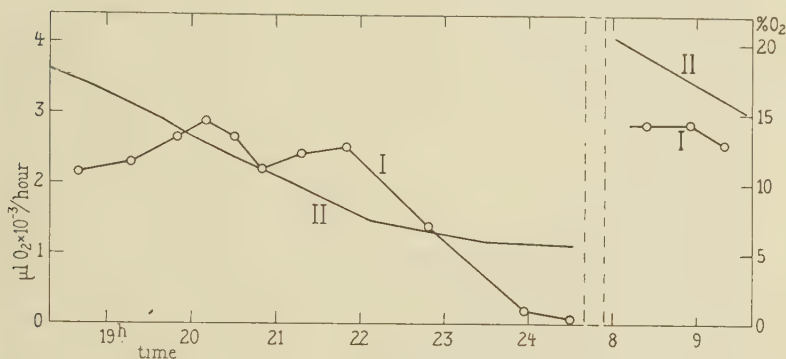


Fig. 14. Trochophora from the plankton (*Phyllodoce*).

Dependency of respiration on average oxygen pressure of the diver. Symbols as in Fig. 13. Between the two dotted lines falls a period of partial, but increasing anaerobiosis.

0.1 μ l diver. Length of animal = 0.39 mm.

the respiration intensity is independent of the oxygen pressure, at least down to 5–6 % of an atmosphere, this being the critical value of the "max. O_2 -pressure in L_2 ". This means that the respiration is independent of the oxygen pressure within the limits normally prevailing in nature. The results obtained here cannot surprise a zoophysiologicalist. Usually, the respiration of small animals is, within wide limits, independent of the oxygen pressure.

As already mentioned previously, lamellibranchs and gastropods are prepared to a periodically anaerobic life within their shells. On veliger larvae of these animals it has been possible through the thin shells to observe that the activity of the cilia can be very low when the animals are retracted into their shells, and the lower, the longer the rest. In these cases, even the foot of the larvae moves very seldom and very slowly. This resting condition is quite temporary, the larva may resume its normal life at any time. In contrast to this, the activity of most nude micro-animals in the plankton

varies only slightly, and the activity of healthy animals will never reach or even approximate zero. Possibly, the special behaviour of the resting mollusc larva has to be ascribed to oxygen deficiency within the shells which contain a continuous CaCO_3 -membrane and, therefore, may be little permeable to O_2 . One single experiment which, however, should not be considered a proof indicates that the

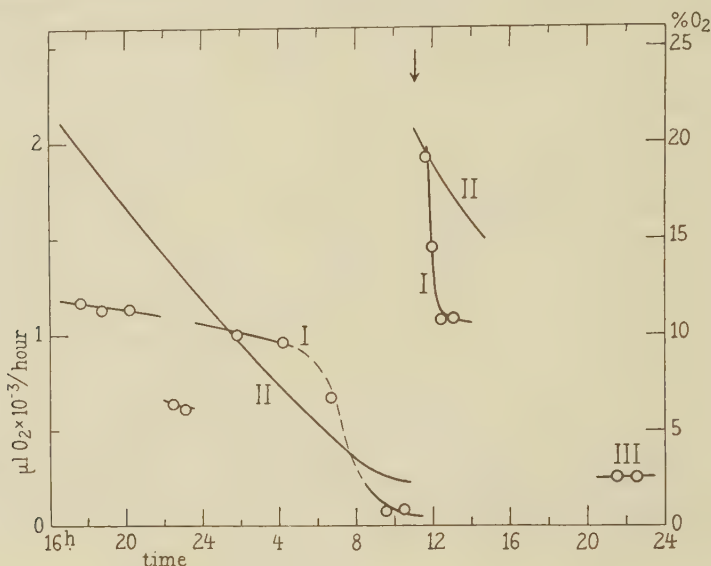


Fig. 15. Lamellibranch veliger larva from the plankton (*Mya arenaria*).

Dependency of respiration on average oxygen pressure of the diver. Symbols as in Fig. 13. Curve III shows the respiration of the isolated velum. The animal swims continuously, except around 23 o'clock.

0.1 μl diver. Length of the animal = 0.22 mm.

respiration of a *Mytilus* larva which was closed during the whole experiment varies with the oxygen pressure over a wide range. Low oxygen tension markedly influences the activity of swimming vermes- and mollusc larvae. This was observed during the experiments published in Figs. 13–15. The observations made on *Pectinaria*, experiment Fig. 13, show that below a certain low limit both respiration and visible activity decrease with decreasing oxygen pressure. At 16²⁰ o'clock, the respiration had decreased to c. 60 % of its initial value as a consequence of a beginning lack of oxygen and, at the same time, the cilia of the trochus were observed to move at a definitely reduced rate. One hour later, the respiration had

decreased to 20–25 % and the cilia in the trochus moved only very slowly. In experiments with *Phyllodoce* (Fig. 14) it was unfortunately unfeasible to observe the cilia. The experiment on *Mya arenaria* (Fig. 15) was exceptional, because there is reason to believe that this larva went on swimming during (almost) the whole experiment, *i. e.* for 22 hours. The larva was not observed between 4³⁰ b. n. and 9⁰⁴ b. n. and just during this period (dotted curve) the metabolic rate and the activity decreased with decreasing oxygen pressure. At 9⁰⁴ o'clock, when the metabolic rate had decreased to 10–15 % of the initial value, the beating frequency of the cilia was enormously reduced. The larva was unable to swim, but it attempted steadily to do so, standing on the bottom of the diver, the velum upwards, but moving only slightly.

In the three experiments, the periods of more or less marked anaerobic life were of different duration, most prolonged in the experiment on *Phyllodoce*, where an eight hours period of marked oxygen lack lies in between the dotted lines. At the end of this period, the animal was at complete rest, only once interrupted by violent muscular movements; the cilia could still not be observed. On *Pectinaria* even the muscular movements were definitely slower than normal. At the time indicated by an arrow, fresh air was introduced into L_3 and L_2 in the following way. The diver was removed from the flotation vessel and the pipette previously used to introduce the M_2 -seal was reintroduced into L_3 , piercing M_2 . Now, atmospheric air was blown into L_3 , and M_2 was placed anew. Finally, the diver was retransferred to the flotation vessel and the readings of the equilibrium pressure were resumed. After the introduction of fresh air, *Mya* and *Pectinaria* in the course of c. 10 minutes gradually regained their normal activity and *Phyllodoce* did so after an even shorter time. Probably, the gradual increase in activity thus observed should be ascribed to a just as gradual increase in the O_2 -pressure in L_2 and in W . When applying the formulae in (118), we find that, in the course of some few minutes, diffusion of O_2 through the seal PO_2 will actually be sufficient to rise the O_2 -pressure in L_2 and in W to such values as will allow the animal in W to resume normal respiration. The respiration intensity, being measured rapidly after the replacement of M_2 , seems to reach or perhaps even to exceed the values to be expected if no oxygen deficiency had occurred. This will be clear from a tentative extrapolation of the—on the whole slightly sloping—curves obtained during the first

part of the experiment. In (118) it is mentioned that a diver charged in the normal way, however without respiring animal, will also change its equilibrium pressure to a not insignificant extent during the "initial period", *i. e.* a period preceding that of the actual measurements. Even after the procedure of introducing fresh air into the diver, an initial period may be present. Thus, the marked oxygen consumption found immediately after anaerobiosis (Figs. 13 and 15) may, in fact, be nothing but the initial period of the instrument and, hence, an experimental error. According to (118) and (42), however, it is typical of the initial period that the equilibrium pressure of the control diver changes at a continuously decreasing rate until the slight change normal for the control divers is reached. In the *Pectinaria* experiment, after introduction of fresh air, the equilibrium pressure changed with an increasing rate, then with a decreasing rate until the respiration was equal to that prior to anaerobiosis. (For technical reasons, the experiment had to be interrupted earlier than desirable). This course of the curve, together with the magnitude of the effects, make the author believe that we actually deal with an oxygen-debt which is paid off under aerobic conditions¹. It should be emphasized that normal activity was reached after the introduction of fresh air in all three experiments before the respiration measurements could be resumed.

The experiments communicated in the present chapter are extremely incomplete, but they quite clearly indicate the respiration of swimming vermes- and mollusc larvae to be independent of the oxygen pressure within the limits normally occurring in nature, *i. e.* the free waters of the ocean².

The larvae can stand such a low oxygen pressure that only a small fraction of their respiration persists, and they respond to oxygen deficiency by a markedly reduced activity. Below a certain O_2 -tension, the activity of the animals decreases with decreasing

¹ Obviously, the experiments do not offer any information concerning the question whether the whole oxygen-debt was paid when the experiments were interrupted.

² In some regions of the sea, for example deep layers of the Baltic and of the Black Sea, a very low oxygen tension may prevail. The experiments indicate that the larvae considered, and maybe most small animals, may penetrate rather far in zones of low oxygen pressure without suffering from lack of oxygen. A similar view was put forward by MARSHALL and NICHOLLS (80) on the basis of experiments with the copepod *Calanus finmarchicus*. Also on the bottom of the sea, the animals may be exposed to low oxygen tensions.

O₂-tension, but not before a very low oxygen tension is reached does the activity cease completely. With increasing oxygen pressure the activity seems again to increase gradually. At normal oxygen pressure the animals resume their normal activity, possibly simultaneously paying off a small oxygen debt. Markedly reduced activity during oxygen deficiency has already been observed by GRAY (31) on the cilia of the *Mytilus* gills.

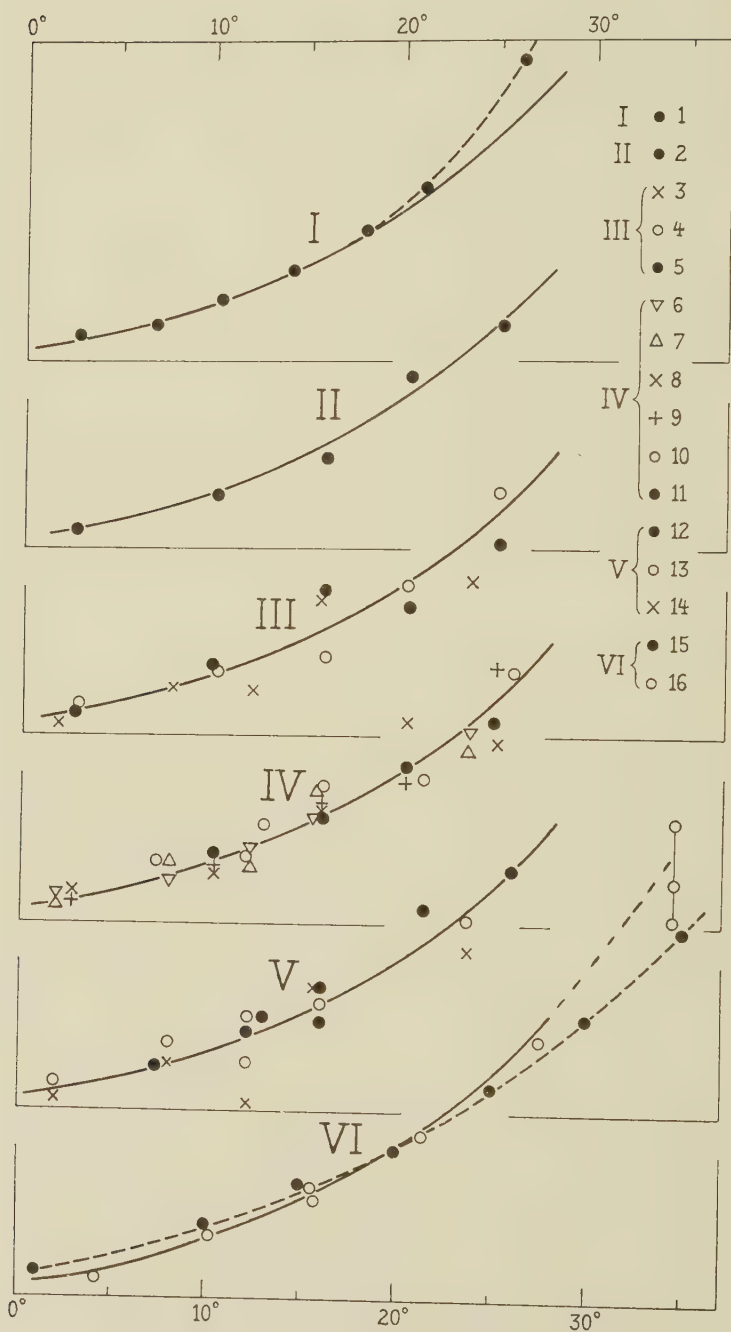
It is known that adult vermes and molluscs, especially from the littoral zone, are prepared to survive periods of complete or partial lack of oxygen, and it must be considered of interest that, in this respect, the larvae can stand the conditions to which they will be exposed later as bottom animals.

It would certainly be of special interest to compare the ability of standing oxygen deficiency of the true plankton organisms (such as *Pleurobrachia*, *Sagitta*) spending their whole life in the plankton, with the organisms studied in this paper, which live in the plankton only part of their lives.

3.13. The Effect of the Temperature on the Metabolism of the Micro-Fauna.

The way how the metabolism of small plankton animals depends on the temperature is worth a study for many reasons. Firstly, by studying such small animals of special living habit, we may extend the material available concerning the temperature-metabolism relationships. Moreover, it can be investigated whether the zooplankton as a whole follows the same metabolism-temperature curve as do the bigger animals. Finally, the metabolism-temperature experiments are valuable because they allow us to make a rough estimate of the minimum work necessary to keep the micro-animals swimming and floating in the plankton (cf. 3.14).

Fig. 16 presents all temperature-metabolism experiments performed. The respiration effect is calculated as the difference between the changes of the equilibrium pressure in the experimental and the control divers, as described in 2.13 and Fig. 3. The effect is not plotted directly against the temperature, but it has been converted in the following way. First, the measured effects are converted to one temperature (0° C) by multiplication with $\frac{273}{273+t}$ (t = temperature of the experiment). In preliminary graphs, the converted



values are now plotted against the temperature of the experiment, and reasonably smooth curves are drawn through the experimental points. Subsequently, the preliminary graphs are comprised in one figure (Fig. 16), using the values of all curves at 20° C as a unit. Fig. 16, therefore, shows only relative measures of the respiration intensity. The fat curve is always KROGH's standard curve (59) and not the average curve drawn through all points. The dotted curves have only been drawn as the best possible ones through the experimental points in those cases in which KROGH's curve was obviously unsatisfactory.

The uppermost curve represents an experiment performed in March 1937 on gemmulae of the fresh water sponge *Spongilla lacustris*. KROGH's (58) micro-respiration apparatus was used. The gemmulac were placed in a very

Fig. 16. Dependence of metabolism on temperature in different micro-animals.

Ordinate: metabolism (arbitrary units). Abscissa: temperature. In all cases, the fat curve is the standard curve of KROGH.

- I. *Spongilla lacustris*.
 - II. *Anthozoa*.
 - III. *Crustacea*.
 - IV. *Vermes*.
 - V. *Mollusca*.
 - VI. Isolated organs of *Mytilus*.
- 1: many *Spongilla lacustris* gemmulae.
 - 2:2 *Anthozo* larvae.
 - 3:49 Nauplii of *Balanus*.
 - 4:1 Unidentified copepod.
 - 5:3 Cypris stage of *Balanus*.
 - 6:16 *Polydora* larvae.
 - 7:6 *Harmothoë* larvae.
 - 8:4 *Harmothoë* larvae.
 - 9:2 *Pectinaria* larvae.
 - 10:1 *Magelone* larvae.
 - 11:10 *Polydora* larvae.
 - 12:10 Unidentified prosobranch larvae.
 - 13:12 *Limapontia* larvae.
 - 14:4 *Montacuta* larvae.
 - 15: Isolated gills of *Mytilus* (GRAY (31)).
 - 16: Isolated velum of a *Mytilus* larva.

For 1, KROGH's apparatus (58) has been used.

For 15, the Warburg apparatus has been used.

For 16, the 0.1 μ l diver was employed.

All other experiments have been performed with μ l divers.

small quantity of water and they occupied a single layer, less than 1 mm thick, on the bottom of a flat vessel which was not shaken. Each experimental temperature was kept constant by $\pm 0.2^\circ$ throughout the experimental period. The curve is drawn according to the above outlined principles, but the dotted part of the curve indicates that the experiments cannot be described quite accurately by KROGH's curve. In the different experiments, the points were gained both with gradually rising and gradually falling temperature. The observation already made by KROGH (59), *viz.* that after a fall in temperature the metabolic rate adjusts itself with a certain inertia, *i. e.* in the course of 15–20 minutes, to the value corresponding to the respective temperature, could also be observed on *Spongilla*.

It thus appears from Fig. 16 that the metabolic rate of 11 different "normally active" micro-animals from the marine summer plankton, and the metabolic rate of one resting organ, *viz.* gemmulae, of *Spongilla lacustris* from fresh water (in March) vary with the temperature in a way hardly deviating from animal to animal. The curves obtained either coincide or at least show considerable similarity with KROGH's standard curve. In view of SPÄRCK's (103) and THORSON's (108) experiments on marine bottom animals it is of importance to state that the veliger larvae of the bottom animals do not seem to deviate from the normal temperature-metabolism relations. The especially low value for the lamellibranch veliger *Montacuta* (cf. Fig. 16) was obtained in a period when the animal was at rest on the bottom of the diver. Thus, in this case, the low metabolic rate may be explained by the low activity which is induced by unknown factors. The isolated velum of *Mytilus* (cf. also 3.144) always swims in the diver and it is deprived of the ability, otherwise typical of lamellibranchs and gastropods, of reacting to different exterior conditions by profound alterations of activity and metabolic rate. Consequently, the metabolism of the isolated velum of *Mytilus* veligers follows almost completely KROGH's standard curve, and the same holds approximately for the gills of the adult *Mytilus* according to GRAY's (31) experiments (cf. GRAY's own curve which is drawn in Fig. 16) and even for the whole intact adult *Mytilus* according to BRUCE (16). There is thus good reason to believe that lamellibranchs and gastropods generally follow rather closely the standard curve, and that SPÄRCK's and THORSON's deviating results must be ascribed to special reactions from the side of the adults. May be, as a reaction towards unphysiologically high or low temperatures, the animals have closed their shells and cut themselves off from the oxygen.

The above material permits a further generalization of KROGH's standard curve which, in first approximation, seems to be valid for numerous lower animals. Higher animals, such as amphibia, mammals (KROGH (59)) and the insect *Dixippus morosus* (BUDDENBROCK and ROHR (referred to in (19))), however, seem to follow a steeper temperature-metabolism curve; perhaps, because on these animals the temperature peripherally affects the metabolism of the tissues and, simultaneously, through the nervous system, affects the muscular tonus and thereby the metabolic rate (KROGH's theory). It may be of interest that insects in diapause follow a very flat temperature-metabolism curve (BODINE and EVANS (8)).

On the basis of the above mentioned experiments it becomes possible from the metabolic rate at a given temperature with reasonable accuracy to predict the metabolic rate also at other temperatures. However, this holds only provided the period of life at the different temperatures is not too long, because we may then expect the setting-in of counter regulations, as discussed in 2.21 and 2.26. For this reason, special experiments are needed in order to decide in how far the metabolic rate measured at a given temperature varies, for instance, with the seasons.

3.14. Experiments on the Floating and Swimming of Micro-Animals in the Plankton.

According to GRAY (31), the work (W) performed by a swimming micro-organism which moves in horizontal direction can be expressed by the formula

$$W = kV^2,$$

while for the same organism the work performed when moving with the same velocity in vertical direction through the water is

$$W = M(\sigma - \varrho)V + kV^2,$$

where V is the velocity of the motion, k is a constant depending on the viscosity of the water, M is the mass of the organism, and $(\sigma - \varrho)$ is the difference between the specific gravity of the organism and that of the water. During horizontal motion, the work kV^2 is necessary to surmount the viscous resistance of the water, for a motion up-

ward in vertical direction also the work of lifting the organism is required, this work being $M(\sigma - \rho)V$.

JENSEN's (45) experiments on *Paramecium* seem to indicate that part of the work of swimming, which is required for the lifting of the organism, is very small as compared with that necessary to overcome the viscous resistance of the water. JENSEN stated the ratio to be 1:10 and GRAY considered this to be rather generally valid, because usually the micro-organisms move at almost the same rate, independent of whether the motion is horizontal or vertical (for exceptions, cf. below). It should, however, be added that for poorer swimmers than *Paramecium* the above ratio must be greater (the size and specific gravity of the organisms considered being the same) and the same holds if the specific gravity of the animals increases. In the case of mollusc veliger larvae carrying heavy shells, the specific gravity of the animals, no doubt, is relatively high as compared with that of water and, according to direct observation, it must also be estimated that the horizontal motion of these animals occurs with a considerably higher velocity than their vertical motion.

In his formulae, GRAY takes into account only the "dynamic work" which is easily defined from a physical point of view and will be denoted as work of swimming. However, GRAY does not take into consideration the work performed in order to keep the organism floating in the water at a constant level; this floating work is difficult to define physically; nevertheless, it requires energy and it must, therefore, be taken into account physiologically.

Until the present, only the work of swimming has been treated in the literature but, in view of the high specific gravity of the veliger larvae, even the work of floating may play a part in these organisms. Every time a veliger larva intrudes the velum, the animal begins immediately to sink at a high rate comparable with or even higher than the rate at which the animal swims in horizontal direction. During horizontal swimming the velum and the body of the animal are kept in a slanting position and we must, therefore, assume that the pull-direction of the velum points slantly upward. If, nevertheless, the direction of the motion is horizontal, this necessarily means that a considerable fraction of the energy production of the velum is required to prevent the animal from sinking, *i. e.* this energy is used in the work of floating. Other animals naturally also yield a work of floating, the magnitude of which is

not very well known. Presumably, it is rather low for animals which do not carry a heavy exterior or interior skeleton.

Until the present, the order of magnitude of the energy required for floating and swimming of the organisms in the plankton has not been touched upon. This question, which is so important from the point of view of comparative physiology, will be treated in the following. We may approach the problem in different ways. Thus, LUDWIG (78) attempted theoretically to calculate the energy exchanges involved in ciliary work and he arrived at extremely low values, the correctness of which the present author feels incompetent to estimate.

3.141. Numerous marine animals reproduce by forming pelagic larvae; at a given stage of their development, however, they stop planktonic life and settle on the bottom at the side of the adults. It will appear from a later section (4.2) that the transition from planktonic to benthonic life does not seem to involve greater changes in the intensity of the oxygen consumption than can be explained by the fact that the plankton larvae are always smaller than the related smallest bottom stages. It will be remembered that the metabolic rate decreases with increasing body size of the animals. The experiments to be discussed later (4.2) on *Asterias rubens*, certain worms, *Nassa reticulata*, *Littorina littorea*, and *Balanus (balanoides?)* thus do not indicate that swimming and floating require considerable additional energy as compared with the life on the bottom.

3.142. It will, moreover, appear from a later section (4.2) that a comparison of different species from the same zoological group and of the same size does not show a systematic difference between the metabolism of plankton animals and that of bottom animals. Just as under 3.141, we may draw the conclusion that neither floating nor swimming requires much additional energy as compared with the benthonic life.

3.143. *The Oxygen Consumption Measured in Different States of Activity of the Single Larva.*

It appeared from section 3.112 that lamellibranch- and gastropod larvae are especially liable to change their activity in the course of the experiment. Thus, in experiments with a single veliger larva

in a 0.1 μ l diver, the animal can be observed sometimes swimming and sometimes moving on the foot or (in the case of bivalves) attached to the bottom, ventilating vividly by means of the gills like an adult animal; most frequently, however, the larva settles down on the bottom of the diver in a state of more or less pronounced rest.

It has been possible at all varying states of activity to measure the metabolic rate and, thereby, to obtain corresponding values of activity and metabolic rate.

The total material indicates that, most frequently, the metabolism increased at the transition from not-swimming to swimming, however, scarcely above twice the lowest value, generally considerably less.

How much does this result tell us about the magnitude of the energy consumption necessary for swimming? If we venture to assume that the activity of the gills, the foot, etc. is unchanged during the transition from not-swimming to swimming, this would mean that the increase in metabolism observed is a direct measure of the energy required for swimming. However, we cannot exclude the possibilities 1) that the larval organs (foot, velum, gills) to some extent take each other's part. In this case, the foot and the gills should be more active in the not-swimming animals than in the swimming animals on which the velum is performing the work. Then, the increase in metabolism measured at the transition to swimming will be smaller than that directly representing the "swimming work" of the velum. On the other hand, not even the reverse possibility can be excluded, *viz.* 2) that swimming involves a general increase in activity of all organs, owing to a general excitation of the animal. Actually, we sometimes see the foot of swimming mollusc larvae move vividly. If the second possibility holds, the increase in metabolism observed at the transition from not-swimming to swimming is too high a measure of the true work of swimming.

Finally, it should be kept in mind that the different states of activity of the animals are rather badly defined. Undoubtedly, the larvae are able to swim, to crawl, and to ventilate with varying intensity and, hence, with varying metabolic rate. Rest (defined as periods when the animal lies on the bottom of the experimental vessel without either swimming, moving the foot, or ventilating) may be of so widely varying character that it seems difficult to decide when the state of rest ends and activity sets in. For example, it is often possible in a plankton sample to point at the resting larvae

which will soon start swimming: these larvae at intervals protrude the velum a little from the shells, unfold it half-way, moving the cilia for a few seconds, only; then, they retract anew, only in order to begin again a little later until, finally, they start swimming. Frequently, the larvae lie quiet for a long period with completely closed shells, performing very rare and weak movements inside the shells. This state may be denoted as deep rest, the metabolic rate being very low; but we cannot exclude the possibility of anaerobiosis because of the closed shells, wherefore no regard will be paid to the experiments on animals which rest so deeply. Since the concept "rest" is difficult to define, the present author often prefers the expression "not-swimming" in order to describe the state of the animals.

It appears from the above discussion that many factors may play a part so that the average increase in the oxygen consumption at the transition from not-swimming to swimming with reservation, only, can be regarded as representing the energy demands of the work of swimming. The observed increase in respiration is not very large, on the average distinctly less than 50 % of the metabolic rate of the swimming larva.

Also some of the experiments with planktonic crustaceans are of interest in this connection. The experiment on the copepod *Centropages hamatus* (Fig. 12) shows that the respiratory rate sometimes decreases continuously over a very long period; this decrease is accompanied—subjectively—by an equally continuous decrease in the visible activity of the animal, so that the metabolism measured at the end of the experiment represents the conditions during the complete standstill of the activity or during seldom and very weak movements of the pleopods. Obviously, it cannot be considered proved that the decrease in metabolic rate and the reduction of visible activity of the animal stand in causal connection; if this is the case, it means that at the most 50 % of the metabolism of *Centropages* is needed for floating and swimming.

3.144. *The Metabolism of the Isolated Velum as Compared with the Metabolism of the Intact Swimming Bivalve Larva.*

In response to violent mechanical stimulation the veliger larva immediately retracts into the shells, and the activity of the cilia of the velum is immediately slowed down. If the larva is not only

stimulated, but injured vigorously, it will react in the same way but, thereafter, it has often been observed that in the course of a few minutes the animal becomes more and more active, the normal swimming inhibition is now lacking and, finally, the animal swims vividly by means of the rags of the velum still left. A whole isolated velum is always extremely active and swims vividly.

The experiments to be described in the following aim at elucidating how much of the metabolism of the swimming larva is located

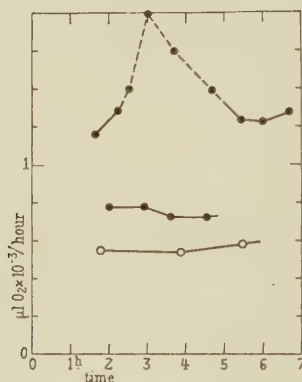


Fig. 17. *Mytilus edulis*.

Upper curve: Metabolism of a single veliger larva. Period of swimming dotted. Middle curve: Metabolism of the isolated velum, measured few hours after the experiment on the intact animal. Lower curve: Metabolism of the isolated velum of an equally large larva. 0.1 μ l divers. Length of animal about 0.25 mm.

in the velum and how much in the rest of the organism. After the respiratory rate of the swimming larva has been measured in the 0.1 μ l diver, the velum is removed by means of needles and, subsequently, the metabolic rate of the isolated velum is determined in another 0.1 μ l diver. In these experiments, we make the assumption—which certainly lends itself to discussion—that the metabolism of the swimming velum is independent of whether the organ is isolated from the animal or not. The velum of bivalve larvae can easily be isolated by pressing the shells together with the aid of a blunt glass rod, while the velum is extruded; hereby the velum is cut off. Another method is to tear the larva to pieces by means of two needles; the velum can then easily be torn off, without being injured, at the stalk connecting it with the animal.

In *Mytilus* less than 50 % of the metabolism of the swimming

larva is located in the velum (cf. Fig. 17), in *Mya* the corresponding figure is circa 25 % (Fig. 15, section 3.12), and in a larva only identified as belonging to the *Teredinidae* the value is found to be c. 15 %. In the last mentioned case, the metabolic rate of the swimming larva was $8 \cdot 10^{-3} \mu\text{l O}_2$ per hour (determined in the μl diver) and the isolated velum consumed only $1.1 \cdot 10^{-3} \mu\text{l O}_2$ per hour (measured in the 0.1 μl diver).

The oxygen consumption per N of the velum and of the whole larva has not been determined; only an attempt has been made to estimate the volume of the velum as compared with the volume of the animal. This estimation favours the assumption that the metabolic rate of the active velum (oxygen consumption/volume) is about twice that of the total larva, but certainly not many times and, possibly, not at all higher than the metabolic rate of the larva.

The fact that the oxygen consumption of the velum of *Mytilus* (as determined in 3 experiments) makes out a higher percentage of the total oxygen consumption than is the case in *Mya* and *Teredinid* can easily be explained by the rough estimate that the velum of the former is relatively larger than the velum of the two latter animals. Perhaps, moreover, the remaining part of the body of *Mytilus* respire especially faintly, as the curve of Fig. 28 p. 96 seems to indicate. This explanation would facilitate the understanding of the relatively high metabolism of the velum of *Mytilus*.

The experiments on *Mya* and *Teredinid* larvae are most important in this connection, because they permit the conclusion (however, obviously limited by the assumptions made in this section) that a velum in which only $1/4 - 1/7$ of the total metabolism of the swimming larva is located will be able to lift and to transport the animal. For this reason, only a minor fraction ($< 1/4 - 1/7$) of the energy consumption at 16° can be required for the work of swimming of the larva. Obviously, these experiments do not tell us whether other micro-organisms need more oxygen in order to perform their work of swimming. An O_2 -consumption corresponding to that found for the lamellibranch larvae described above will presumably be sufficient to keep the organism floating and also swimming at a moderate rate. Naturally, more rapid motions will require more energy.

3.145. *Swimming with Reduced Metabolic Rate owing to Low Temperature.*

From all observations discussed above (3.141–3.144) we may draw the conclusion that presumably considerably less than 50 % of the oxygen consumption of the small planktonic animals (at 16°) are needed for the work of floating and swimming. As long as we consider the possible large changes of the oxygen consumption which might occur during sedimentation and metamorphosis, the above mentioned 50 % must be regarded as a relatively small value; however, as an absolute measure of the work of swimming, this value is not insignificant, because the metabolism of the micro-organisms is so relatively high. In the present section, some arguments will be brought forth in favour of the view that the above mentioned value of 50 % presumably is much too high.

It is of importance in this connection to notify that the respiratory rate of the isolated velum of the *Mytilus* larva showed the same temperature dependence as did the respiratory rate of the whole animal. On the supposition that the same holds for the velum *in situ* and, moreover, that the mechanical efficiency of the metabolism is almost independent of the temperature, it becomes clear that, at low temperature, the velum yields only very small amounts of energy for the swimming. Generally, we must imagine that the locomotory organs of other animals behave analogously at decreasing temperature. Will organisms normally living in the summer plankton (16°) then altogether be able to swim in water of the winter temperature of 2–3° or even less, or do they sediment at such low temperatures? Orientating experiments concerning this question were performed at the end of the summer of 1944; frequently, it could be observed that the animals were less active at low temperature than at higher temperatures. However, at the prevailing temperature, all animals were able to rise for swimming. The temperatures varied between 1.5° and 24°, and the following animals were investigated:

<i>Mytilus</i>	}	lamellibranchs
above discussed <i>Teredinid</i>		
unidentified mollusc I		
unidentified mollusc II		

<i>Littorina</i>	}	gastropods
<i>Limapontia</i>		
Balannauplii	}	crustaceans
various copepods		

The oxygen consumption at 1.5° of a plankton organism or of an isolated velum amounts only to circa $\frac{1}{6}$ of the oxygen consumption at 16° . The energy consumption (at 16°) of the isolated velum of *Mytilus* was about 50 % of the energy consumption of the whole animal at the same temperature. Therefore, the energy necessary to keep this animal floating or in moderate motion in water (at 1.5°) amounts at the highest to $50\% \cdot \frac{1}{6} = 8\%$ of the energy production of the organism at 16° C. In the case of the *Teredinid* an even lower figure may be adopted, namely circa $15\% \cdot \frac{1}{6} = c. 3\%$ of the energy production at 16° C. As mentioned earlier, the mechanical efficiency of the metabolism, viz. $\frac{\text{work performed}}{\text{energy consumed}}$ was assumed constant at all physiological temperatures. The higher viscosity of water at $2-3^{\circ}$ $\left(\frac{\eta'}{\eta} = 1.6-1.7\right)$ as compared with the viscosity at 16° $\left(\frac{\eta'}{\eta} = 1.1\right)$ may play a part in changing the efficiency, however hardly to such an extent as to falsify the above conclusions.

Hence, according to the present experiments, the energy necessary for floating and swimming at moderate rate is only a small fraction of the total energy consumption of the organism. Presumably, for all or for the majority of the micro-plankton animals, the energy necessary for floating alone (definition above) is negligible as compared with the total energy consumption of the animal. This conclusion may certainly be generalized, as it is based mainly on experiments with the relatively heavy veliger larvae whose work of floating, as mentioned before, must be comparatively great.

As regards the work of swimming, we can only say that low or moderate activity, as observed on the micro-animals of the plankton at $1-2^{\circ}$, probably only requires a very small fraction of the total energy consumption of the animal. On a number of organisms moving by means of cilia—for example asteride and trochopore larvae—the ciliary system is rather less developed than that

of the veliger larvae discussed above. Therefore, it must be considered probable that even at 16° at the most a few per cent of the metabolism of these animals is required for floating and swimming. However, on the basis of the present experiments, we cannot exclude the possibility that swimming at a higher rate, as it is observed on certain organisms at 16°, requires up to 50 % of the total energy production, although a lower figure appears more probable.

3.146. *Conclusion.* From what is said in sections 3.141–3.145 we may draw the conclusion that, presumably, the work of floating and swimming of the micro-animals of the plankton requires less than 50 %, in some cases perhaps only a negligible fraction, of the total energy production of the animals. The work of floating alone requires very little energy. This renders it justified in the following to compare the author's own measurements of the metabolic rate of micro-animals with those on larger, mostly resting animals to be found in the literature.

4. THEORETICAL PART

ON THE RELATIONSHIP BETWEEN BODY SIZE OF THE ANIMALS AND THEIR METABOLIC RATE

4.1. *Introduction.*

The following survey cannot possibly comprise more than a minor fraction of the comprehensive literature dealing with the relationship between the body size (weight) and the metabolism of the animals. The author will, therefore, discuss only what is regarded as indispensable to give a rather complete picture of the problem. In KROGH's book "The Respiratory Exchange of Animals and Man" the reader will find a good deal of the literature up to 1916. The more recent literature is very scattered and—as far as it is known to the present author—has nowhere been collected completely. We must, therefore, confine ourselves to BUDDENBROCK's (19) and BENEDICT's (5) books. After the conclusion of the present work the author received BRODY's comprehensive and most important book (14) which contains a treatment of the extensive and spread literature concerning the metabolism of domestic and other large homoeotherm

animals. Only in a few cases where it was considered absolutely necessary, reference has been made to BRODY's book after the present manuscript was finished. No comparative investigation of the metabolism/N of the animals seems to exist; the experiments described in the present work are the first attempts of that kind.

In the literature, the animals are compared either intraspecifically (animals of the same species, but of different size) or interspecifically

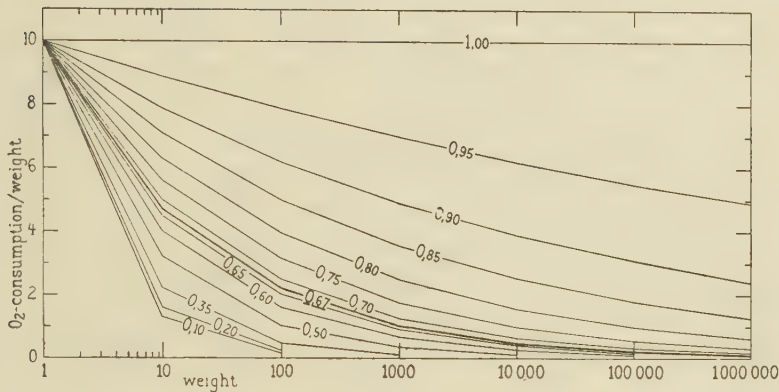


Fig. 18. Diagram, see text.

(animals of different species and size). It remains obscure whether the results obtained depend systematically upon the basis of comparison; this, however, will later be discussed in greater detail. The author's own experiments on worms and crustaceans seem to indicate that, within larger groups of animals comprising species of greatly varying body size (variation $> 10^4 - 10^5$ times), the size factor is of dominant importance for the metabolism of the animals. For this reason, the curves depicted in the following show the experimental results plotted in a coordinate system where the abscissae represent the weight or the N-content of the animals, and the ordinates show their metabolism calculated per unit of N or per unit of wet weight. In the present work, metabolism per N or per weight is denoted as metabolic (respiratory) rate. The ordinates are equidistant, while the abscissae are in a logarithmic scale, whereby it is possible to compare in one graph animals of widely different size. Although factors other than body size (growth rate, age, activity, etc.) thereby are neglected because they are regarded as being of secondary significance, these factors may possibly be the cause of

the marked scattering of the points around the average curves drawn.

Let us consider an animal of the body size 1 and having the metabolic rate 100. If this animal is compared with related animals of the sizes 10, 100, and so on, we may read from Fig. 18 which metabolic rate the larger animals would have, provided their meta-

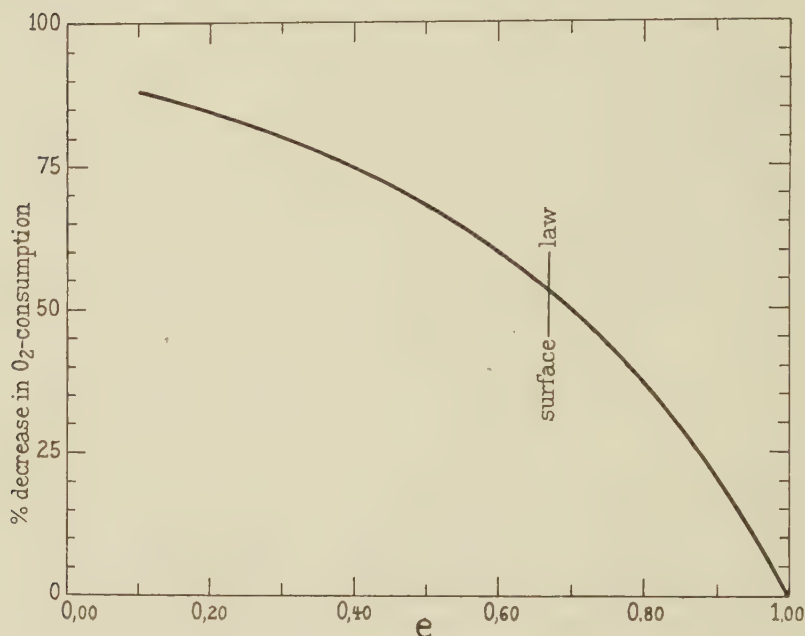


Fig. 19. The relation between e and the "percentage decrease in metabolic rate at a 10-fold magnification of the animals".

bolism was proportional to W^e , where e is varied in the figure. The fat curve represents the value $e = 0.67$ and corresponds to RUBNER's surface law. Evidently, a comparison of animals within a narrow size variation requires very exact experiments in order that a reasonably accurate determination of the value of e may be reached. Here, we encounter one reason for the confusion prevailing with regard to the value of e . In view of the fact that, in most cases, e has only been determined very inaccurately, we may realize that any attempt of calculating by way of extrapolation (cf. PÜTTER (93)) from experiments on animals of some intermediate size the metabolic rate of very small and of very large animals may lead

to serious errors, especially because it is by no means proved that the value of e does not change with the size of the animals considered (cf. Section 4.4).

The danger involved in such extrapolations may be illustrated by Fig. 18. If e is equal to 1, we find the same metabolic rate for all animals, irrespective of the weight. If e is 0.85, however, we find the ratio between the metabolic rate of animals of the weight 1 and of animals of the weight 10^6 to be 8.7. In the case of $e = 0.67$, finally, we find the same ratio to be 82.

In the analysis of the experimental curves the author has abandoned the term e and, instead, made use of the term "the percentage decrease in metabolic rate at a ten times magnification of the animals." Each exponential function $y = W^e$ is characterized by the fact that to a given value of e corresponds a given value of the "percentage decrease in metabolic rate at a ten times magnification of the animals." The interdependence between e and the "percentage decrease" is represented in Fig. 19. Thus, this figure may be used if the results published in the present work have to be converted into e -values.

4.2. Metabolism/N of Marine Animals in Relation to their Size (N-content), with Special Regard to the Micro-Fauna.

The experimental figures concerning the respiration/N of the animals are only of little value when considered isolated. Together, they permit a comparison of the metabolic intensity of animals of largely varying body size.

The present chapter comprises in a series of graphs the author's own experiments on different marine animals. The signatures are explained below the figures. It may only be mentioned here that two or a few points connected by a line parallel to the axis of ordinates represent several respiration measurements on one and the same animal, divided by the N-content of the animal. The average curves may frequently look rather arbitrary; they have only been drawn in order to facilitate in a later chapter of this work a rough comparison of different groups of animals.

4.21. Coelenterata.

A few experiments were performed on *Sarsia* sp. (Fig. 20), on one young jelly fish, *Aurelia aurita*, and on *Pleurobrachia*. Most animals were 1–2 mm in diameter and, except for one single large *Pleurobrachia*, weighings could not be carried out. On this single animal, the respiration/weight was found to be very low, but the

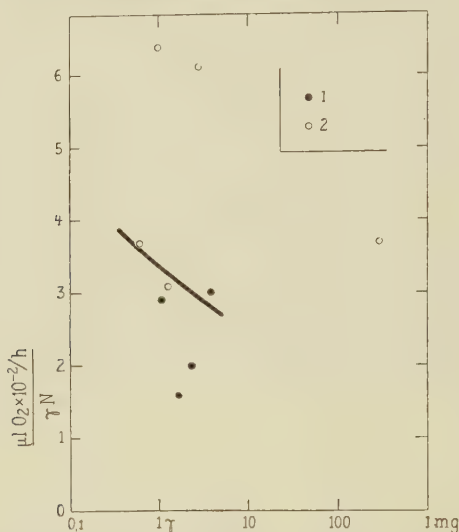


Fig. 20. *Coelenterata* from the plankton. Metabolism/N (ordinate) and body-N (abscissa).

1. Different jellyfishes.

2. *Pleurobrachia*.

All animals (except the largest one) are 1–3 mm in diameter.

respiration/N is not markedly different from that found in other lower animals. It is not surprising that *Pleurobrachia*, which seems so very active as compared with *Sarsia*, also respire more intensely.

Thus, the N of the *Coelenterata* appears to be metabolically just as "active" as is the N of many other animals of the same size. However, it should be emphasized that, in these animals, both respiration and N may mainly be localized to less jellified parts of the body (marginal sense organs, muscles, swimming plates, etc.).

4.22. Echinodermata.

From the echinoderms, only the starfish *Asterias rubens* is included in the present investigation. *Asterias* is a bottom animal

forming pelagic, relatively large larvae. During metamorphosis the larvae settle to the bottom where they may be found as young starfishes, circa 1 mm in diameter.

Fig. 21 represents the experimental results. Large bottom animals (wet weight 5–25 g) respire much less intensely (only c. $1/5$ – $1/10$) than do the plankton larvae which are 10^6 times smaller. However, it becomes clear that the change from life in the plankton to life on the bottom is not the decisive factor, since very small,

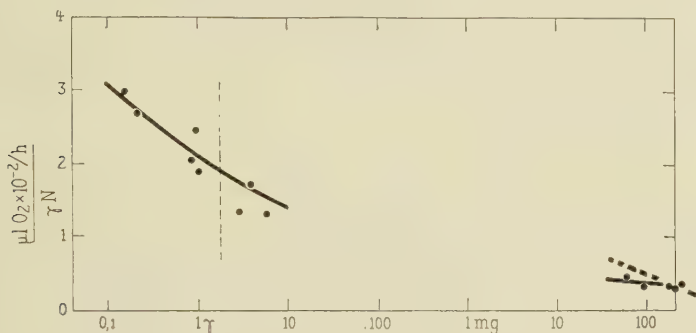


Fig. 21. *Asterias rubens*.

Metabolism/N (ordinate) and body-N (abscissa).

The dotted line parallel to the ordinate indicates the limit between planktonic and benthonic life.

The dotted curve to the right describes the experiments of MEYER.

just sedimented individuals have almost the same oxygen consumption as have the plankton larvae (the vertical dotted line of Fig. 21 indicates the size of the animals when sedimentation occurs).

There may be some reason to assume that no other change in the oxygen consumption of *Asterias* occurs on sedimentation and metamorphosis than what may be explained exclusively by the simultaneous change in size of the animal. The same seems to hold for all other animals studied, even if the demonstration is in some cases very uncertain.

It appears from HELGA MEYER's work (81) that the oxygen consumption of adult *Asterias* is rather difficult to measure because, within wide limits, it increases with increasing oxygen tension in the water. This is due to the fact that *Asterias* because of a poor blood circulation is largely dependent on the oxygen which is transported to the tissues by diffusion. For adult *Asterias*, MEYER found the relation illustrated by Fig. 33 between weight and respiratory

rate. The steepness of the curve may possibly be explained by the assumption that the oxygen diffusion into the animal is the more difficult the larger the animal.

If we assume MEYER's animals to contain 1 % N, the average found for the Helsingør animals, we may from her experiments calculate a curve representing respiration/N. This curve is roughly indicated by the dotted line of Fig. 21. The present author has greater confidence in MEYER's careful experiments than in his own few ones and he, therefore, considers it possible that further experiments will show a very slightly sloping curve in the empty middle region of Fig. 21, but a steep decrease when the animals approach their largest possible body size.

4.23. Vermes.

Fig. 22 shows the respiratory rate in relation to the body size of different worms. Except for *Polydora ciliata* to be found in Fig. 23, the figure represents all the present author's experiments on *Vermes*. One point in the figure refers to the geophyrean *Phoronis*, two refer to experiments on nematods from the sand bottom at a depth of 2 m near Snekkersten, all other points represent experiments on different polychaete worms. On an average, the smallest larvae respire c. 4 times more intensely than do the largest bottom animals studied, the latter being c. 10^5 times larger than the former. Considering the figure from the right side, we meet with adult bottom stages of Nereids, *Arenicola*, *Lepidonotus*, *Fabricia*, "polychaete worms from Snekkersten", and nematods. The rest of the points refer to larval planktonic worms. All observations are intimately intermingled so that the decrease in the metabolic rate of the animals seems to be related neither to the habit of living nor to the ontogenetic stage of development of the respective animals. The curve, therefore, may be considered to represent the true relation between body size and metabolic rate. The fact that all experimental animals suit the system can be taken to indicate that this relation holds for the order *Polychaeta* as a whole, and there is some indication that it even holds for the phylum *Vermes*. Obviously, from this relation we cannot predict whether future experiments on a different species will yield results which will agree with the system shown in Fig. 22. Possibly, or even probably, experiments on other species, especially those deviating considerably from the species

used here with regard to morphology, visible activity and living habit may lead to results which are essentially different from those depicted above. Experiments on *Vermes* from very warm or very

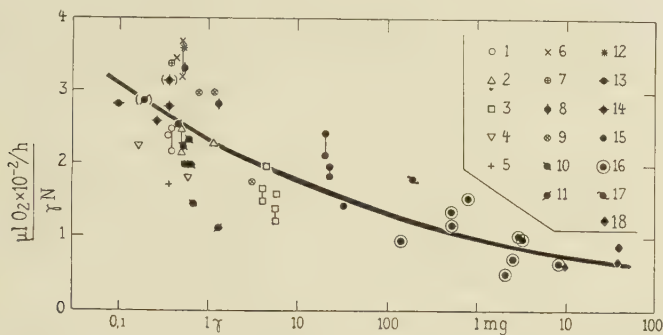


Fig. 22. *Vermes*.

Metabolism/N (ordinate) and body-N (abscissa).

1. Nematods from sand bottom, off Snekkersten. Depth 2 m.
2. *Capitella* sp., adult.
3. *Fabricia* sp., adult.
4. } Different *Phyllodoce* larvae from plankton.
5. }
6. }
7. *Phyllodoce* (*groenlandica*?) larva from plankton.
8. *Nephtys* larva from plankton
9. *Magelona* from plankton, larval.
10. *Disoma* from plankton, larval.
11. *Harmothoe*, larva from plankton, adult from bottom. The experiment in brackets is uncertain.
12. *Phoronis*, larva from plankton.
13. Unidentified larva from plankton.
14. *Pectinaria* larvae from plankton, adult from bottom.
15. *Polydora*, adult.
16. *Nereis*, adult.
17. *Lepidonotus*, adult from bottom.

cold climates and, moreover, experiments on various endoparasites, will undoubtedly offer interesting informations.

Unfortunately, the experiments on *Polydora ciliata* which during the work was selected to be the main object of study among the *Vermes*, turned out to be the poorest of all. *Polydora* often appeared injured in the diver, viz. the setae were cast and the body was locally cytolized. The reason for this may be that the animal reacts very characteristically upon touching: it spreads out the setae and,

thereby, makes pipetting rather difficult and, maybe, injurious. The difficulties of pipetting the animals were met with again when they had to be removed from the diver and, moreover, when they were transferred to the ampulla prior to the N-determination. Thereby, we introduce uncertainties both of the respiration measurement and of the N-determination. The high scattering of the points of Fig. 23

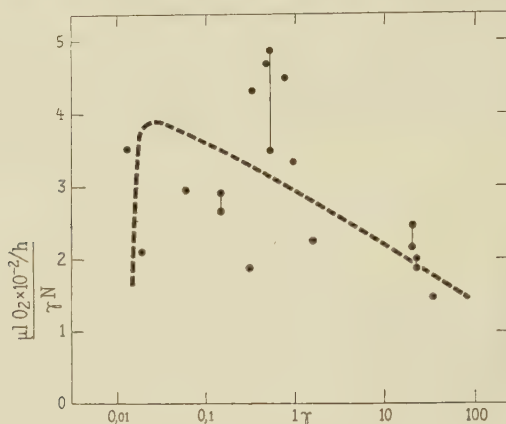


Fig. 23. *Polydora ciliata*.

Relation between metabolism/N (ordinate) and body size, expressed by the N-content (abscissa). The two points to the left represent experiments on embryos removed from the mother animal. The points to the right represent experiments on adults from *Littorina* shells. All other points are experiments with larvae from the plankton. The curve is drawn analogously to Figs. 25, 26 and 28.

must be considered due to these circumstances. Even if the results of Fig. 23 are not in contrast to those of Fig. 22, it was found convenient only to include in the latter figure the experiments on adults of *Polydora* which were easy to handle.

4.24. Mollusca.

In the case of *Asterias* and *Vermes*, the activity of the animals under investigation was low and did not seem to vary very much from one species to another. Moreover, these animals appeared rather unaffected by the experimental conditions. The *Mollusca*, however, behave differently: usually, these animals are equipped with shells which most probably are almost impermeable to oxygen, and the animals may retract into these shells as soon as the outside

conditions do not suit them. Frequently, therefore, they may more or less completely cut themselves off from the oxygen and for some time live anaerobically. This holds especially for the larger bivalves which are provided with a thick calcareous shell; but even the shells of mollusc veliger larvae seem to prevent effectively the diffusion-in

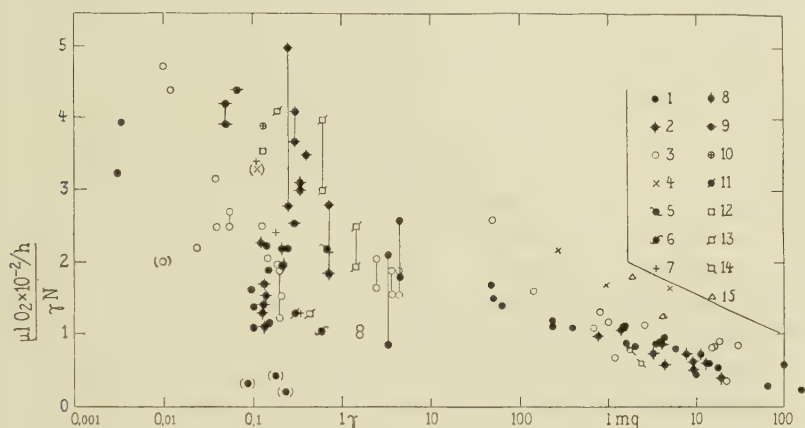


Fig. 24. All experiments with molluscs.

Relation between metabolism/N (ordinate) and body size, expressed by the N-content (abscissa).

1. *Mytilus edulis*, adult and larva.
2. *Nassa reticulata*, adult and larva.
3. *Littorina littorea*, adult and larva.
4. *Cardium edule*, just sedimented.
5. Unidentified snail larva.
6. Unidentified snail larva.
7. *Limapontia*, larva.
8. Unidentified lamellibranch larva.
9. *Cerithium* larva.
10. *Montacuta* larva.
11. Unidentified gastropod larva.
12. *Zirphaea* larva.
13. Unidentified gastropod larva.
14. *Macoma baltica*, adult.
15. *Mya arenaria*, adult.

of oxygen. Thus, it is easy to understand that the oxygen consumption of many molluscs may be very variable and difficult to measure in a state of "natural activity", *i. e.* in the case of the veliger larvae, swimming, and in the case of the bottom forms, crawling or ventilating normally.

In Fig. 24, all experiments (in the μ l diver and with Winkler titrations) on bivalves and gastropods are pictured. The metabolism of the bottom animals was determined by Winkler titrations, and great stress was laid upon noticing the activity of the animals during the experimental period. Experiments on passive animals have generally been discarded. The only exceptions from this rule are the three experiments (in brackets) on *Mytilus* larvae and perhaps one on a very young, just metamorphosed *Cardium*. Finally, one less accurate experiment with *Littorina* larvae (removed from a cocoon) is given in parenthesis. All respiration experiments on veliger larvae were carried out in the divers. In most cases, some few animals (5–20) had to be used for an experiment, partly in order to obtain a measurable O_2 -consumption, partly in order to get a sufficiently large quantity of N for the analysis. However, most of the animals were obviously less liable to swim in the divers than when they were free to move in larger volumes of water. In spite of the fact that prior to the experiments all animals were beautifully swimming, usually only 10–30 % (sometimes fewer) of the animals did actually swim in the divers. The respiration measurements, therefore, can only yield results which—when calculated for one single larva—must lie somewhere between the oxygen consumption during swimming and that during rest. The deviation from the true value—the metabolism of swimming—will depend on how much the metabolic rate changes at the transition from rest to swimming, or *vice versa*.

All N-determinations given in the figure represent the total N-content of the animals. Obviously, we wish to compare the respiration of the animals on the basis of the N-content of the tissues, only. In the case of larvae, however, only the total N-content could be accurately determined. Imperfect attempts to distinguish between the N-content of the shells and that of the tissues indicate that no more than 10 % of the N of the larvae is located in the shells (*Mytilus* and *Nassa*, Fig. 27). On adult specimens of *Nassa*, *Littorina*, *Cardium*, and *Macoma* still less was found in the shells, while in *Mytilus* c. 35 % of the N of the adult animal was present in the shells. Hence, these experiments are in favour of the view that Fig. 24 gives a relatively good impression of the relation between body size and metabolic rate of the molluscs studied, even when the comparison is performed on the basis of N in the tissues. Not even in the case of *Mytilus* would a correction for the N-content of the shells lead to an essential change of the picture. Obviously, the highest

weight should be given to those experiments in which most larvae actually do swim and, consequently, the metabolic rate is highest. For this reason the experiments on *Nassa* larvae are of the greatest value. It seems impossible to draw a reasonable curve through the swarm of points. Marked differences between different species seem to exist, and this is the reason why in the following more attention should be paid to the three species *Nassa reticulata*, *Littorina littorea*, and *Mytilus edulis*.

4.24I. *Nassa reticulata*.

Fig. 25 represents all experiments performed on larvae and adults of *Nassa reticulata*. From left to right, we have first a few experiments on embryos (*a*) removed from the cocoon and still containing yolk, all swimming slowly. Respiration/N is low, presumably because not negligible amounts of "respiratory inactive" N are situated in the yolk. During this early stage of embryonic life the N of the embryo remains constant, but the respiration increases (the curve is drawn vertically through the points). This may be due to the conversion of the yolk into respiring tissue protein. Then follows (*b*) an experiment on three larvae which swim at the start of the experiment (highest point) and are at rest at the end of the experiment (lowest point). *c* represents an experiment on three swimming larvae (lowest point). In the course of the measurements, one or maybe two or three larvae metamorphosed (indicated by the presence after the experiment of numerous isolated velum cilia in the diver). The following morning, a new experiment (highest point) is started on the same larvae which now crawl lively. The metabolism is slightly higher, at any rate not lower than prior to the metamorphosis. *d* and *e* are experiments on five larvae in each diver. Direct observations in the diver were difficult, but at any rate 2-3, and possibly all larvae were swimming in both experiments. Finally, *f* is an experiment on an exceptionally large veliger larva of *Nassa*. At the start of the measurements, the larva swam and the respiration intensity was high; at the end of the experiment, the larva was crawling and its respiration was lower. A detailed discussion of the experiments *b* and *f* may be found in 3.11 and in Figs. 10-11. All experiments of Fig. 25 are calculated as respiration/total N. The experiments on adult *Nassa* from the bottom are given to the right. Also these experiments are expressed as respiration/total N. How-

ever, only about 10 % of the total N of the larvae are contained in the shells (cf. Table 2, Section 2.163) and the shells of the adults contain still less (unpublished experiments). Thus, almost the same relation as that in Fig. 25 would have been found if the results had been expressed as respiration per N of the tissue. Moreover, the respiration was measured on animals which were "normally active". The curve of Fig. 25 must, therefore, be considered to represent

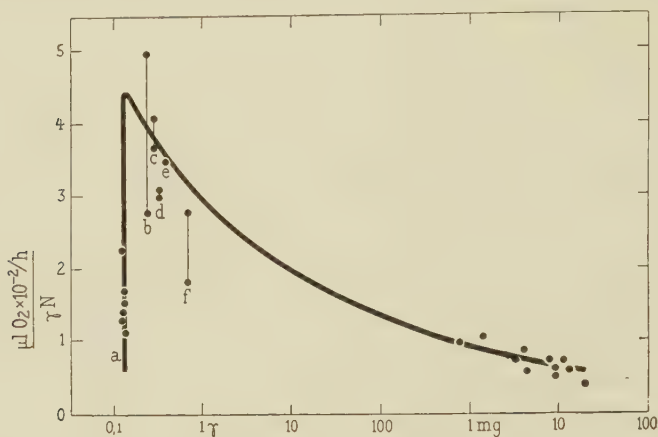


Fig. 25. *Nassa reticulata*.

Relation between metabolism/N (ordinate) and body size, expressed by the N-content (abscissa).

a true relation between the metabolic rate of adult, crawling bottom animals and that of swimming veliger larvae. In other words, this is the relation between the metabolic rate of small and of c. 10^4 times larger animals of the same species, all studied in "normal" activity. The difference between the metabolic rate of swimming and of crawling larvae seems not to be very marked (cf. also Figs. 10–11), and metamorphosis is probably not connected with any essential change of the respiration intensity. The metabolism/N of the larva is about 4–5 times higher than that of the adult.

4.242. *Littorina littorea*.

We shall first confine ourselves to the discussion of the experiments marked with small circles in Fig. 26. They were performed on many veliger larvae (to the left of the dotted line) or a few very

young bottom animals in the μl diver or, finally, on a few or single older bottom animals ($N > 10\gamma$) in Winkler experiments. In order to obtain a sufficiently high respiration effect and a quantity of N sufficient for the analysis, it was necessary to use a few or several veliger larvae in each diver experiment. However, with increasing body size the *Littorina* larvae are less and less liable to swim in the diver; thus, only in the experiments *a* and *b* performed on larvae

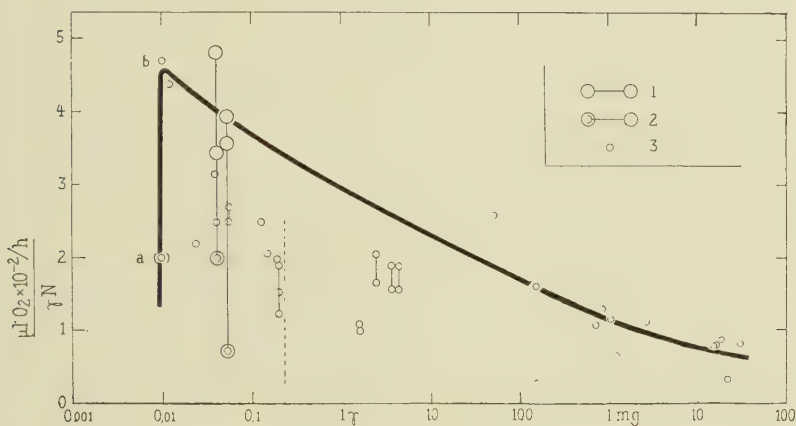


Fig. 26. *Littorina littorea*.

Relation between metabolism/N (ordinate) and body size, expressed by the N-content (abscissa). The point in brackets is uncertain.

just removed from the cocoon (*a*) or just hatched (*b*), all or almost all larvae swam during the experiment. In all other experiments on *Littorina* larvae, less than 30 % of the animals swam, and it must therefore be concluded that the values obtained are to an unknown extent lower than the swimming metabolism to be measured.

It is known from experiments on *Nassa* (cf. 4.241) that the respiration/N of the newly hatched larva need not be a true expression of the respiration/N of somewhat older larvae the yolk of which has been consumed and which grow on nutrition taken up from outside. We must, therefore, attempt on the basis of special experiments to obtain some information on the metabolic rate of such slightly larger, swimming *Littorina* larvae. These experiments were performed in the 0.1 μl diver.

Only one *Littorina* larva was used in each experiment. The method is sufficiently sensitive to record the metabolic rate of this

larva even during very short periods of, for example, 10 minutes. Thus, the variation of the metabolic rate as a function of the activity of the animal can be followed. In experiment *A*, Fig. 9 of Section 3.112, we start with a long period of swimming, the metabolism being high, but not constant; this may indicate that the animal can swim with varying intensity. This manifests itself by varying metabolic intensities of the animal. After swimming comes a period of rest during which the metabolism is again markedly reduced. At the end of the experiment the animal again swims, and the metabolism is increased. In the present experiment, the transition from the condition of rest to that of swimming involves at the most a doubling of the metabolism. The result of experiment *B* is somewhat different. During the first 8–9 hours, the larva is at rest and the metabolism decreases continuously. At 23 o'clock, the larva starts swimming and the metabolism is increased by a factor 6. Not too much stress should be laid upon the fact that the metabolic rate is so low during the period of rest, since this may be due to anaerobiosis in the animal whose shell was probably closed. It is of greater importance to determine the metabolism of the swimming larva, and this measurement could be carried through successfully in both cases.

However, the metabolism measurements represented in Fig. 9 cannot be converted into measurements of metabolic rates, unless we have gained a measure of the N-content of the respective larvae, which latter value was too low for a direct determination.

In Fig. 27, the N-content of the *Littorina* larvae used in the main experiments of Fig. 26 is plotted as a function of the largest shell diameter. Using this curve and the largest shell diameters of the above larvae *A* and *B*, we may calculate their N-contents and, thus, also their metabolic rates (respiration/N) both during swimming and during rest. The values arrived at in this way are given in large circles in Fig. 26. For each experiment, three points connected by vertical lines are shown. The two highest points of each experiment represent the highest and the lowest metabolic rate, respectively, measured during swimming, while the lowest point (with two circles) shows the lowest metabolic rate at rest. Thus, as was to be expected, the metabolic rate during swimming is somewhat higher than what was found in the experiments on numerous larvae in the μ l diver (small circles). This is in agreement with the contention that the metabolic rate of the swimming *Littorina* larva is higher

than that of the resting larva (Fig. 9). All experiments of Fig. 26 are expressed as respiration/total N, but probably the same general trend of the curve would have been found in case we had compared the experiments on the basis of N in the tissues, only. The N-content of the shells of these larvae could not be determined, but most pro-

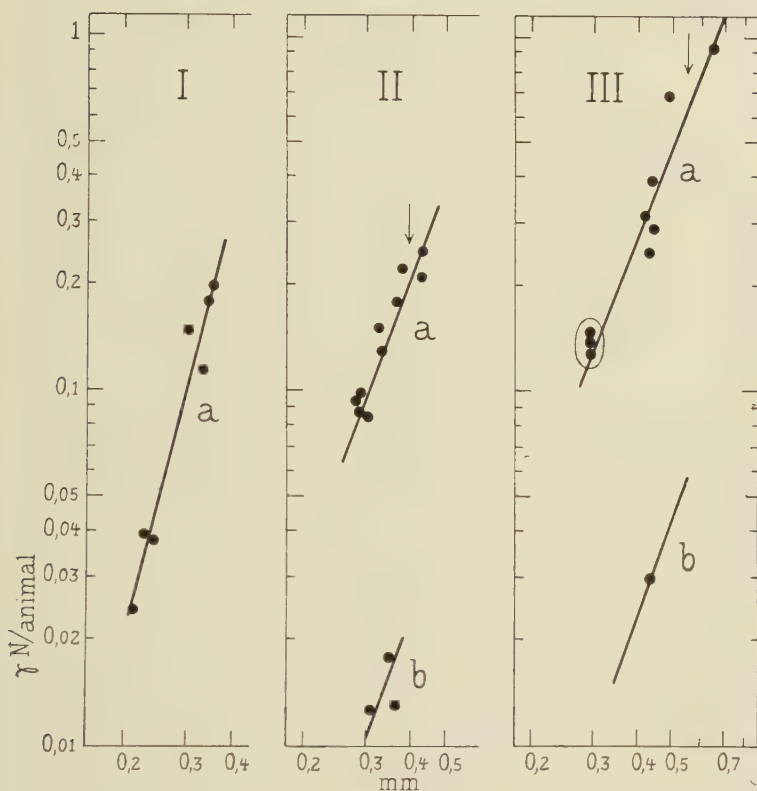


Fig. 27.

a curves: Relation between largest shell diameter (abscissa) and total N-content (ordinate) in the larvae of

I. *Littorina littorea*

II. *Mytilus edulis*

III. *Nassa reticulata*.

The points to the left of the arrow indicate experiments with planktonic larvae; the points to the right of the arrow denote experiments on young bottom stages. The encircled points in III represent experiments with embryos removed from the cocoon.

b curves: Experiments with isolated shells.

NB. The double logarithmic coordinate system.

bably it is very low, just as in *Nassa* and *Mytilus* (cf. Fig. 27). In the case of adult specimens of *Littorina*, the N-content of the shells has generally been found to be considerably below 10 % of the total N of the animals. The curve in Fig. 26 was drawn with special regard to the above described experiments on single individuals, and it thus illustrates the relation between body size and metabolic rate of *Littorina* during "normal" activity, in so far as this state of activity can altogether be defined. Hence, plankton larvae of *Littorina* respire about 4–5 times more intensely than do the 10^5 times larger bottom animals.

It remains undecided whether metamorphosis is accompanied by a slight decrease in metabolic rate or not. We must confine ourselves to the statement that, in *Littorina*, sedimentation and metamorphosis do not involve any essential change of the metabolic rate.

4.243. *Mytilus edulis*.

Fig. 28 shows the metabolic rate (metabolism/total N) of *Mytilus edulis* plotted against the size of the animal, as expressed by its total N-content. Also in this animal, the metabolic rate decreases with increasing body size, however, definitely less than in many other animals. Moreover, the shape of the curve is different from that found for other species.

It is quite obvious that the points of Fig. 28 show a scattering markedly exceeding that obtained in experiments on other animals, for example *Asterias rubens* (Fig. 21). The fact that the scattering of the results obtained varies from test animal to test animal seems to indicate that the scattering is not due to experimental errors, but to biological variations (cf. also 2.1, the discussion of the accuracy of the technique applied). It will, moreover, appear from the following that the experimental conditions affected the activity and the metabolic rate of the animals and, thus, contributed to increase the biological variation. These circumstances made it necessary to vary the technique applied in order that the metabolic rate of the animals could be measured, as far as possible, under "normal activity".

On the left of Fig. 28 we meet with the two first and highest points of the figure, representing respiration/N of very young trochophorae developed from eggs which were fertilized in the laboratory 24 hours earlier. All of them swim vividly; they are very small and,

within a few hours after this measurement, they have already developed into young veliger larvae. The vertical line on the left side of Fig. 28 indicates that the respiration intensity of these young embryos is supposed to be the result of an increase in respiration intensity occurring after fertilization. Because so few measurements are available, we cannot regard it as certain that the latter increase in respiration intensity would not even surpass the values depicted in Fig. 28.

The next measurements given in Fig. 28 (going from left to right) were performed on young veliger larvae from the plankton. (At the moment, we disregard the points surrounded by circles, which will be discussed later.) The experiments were carried out on numerous larvae in each μ l diver. The larvae in the divers, however, behaved somewhat different from experiment to experiment, dependent on unknown factors playing a part in the experiments. The lowest respiration intensities were found in experiments (1-7-8 of Fig. 28) where all, or almost all, larvae lie quietly on the bottom of the diver, their shells being closed or at least very slightly and rarely opened. Closer inspection of the animals after the conclusion of the experiments, however, showed that the animals were alive and healthy; occasionally they started swimming. These few experiments have not been taken into account when drawing the curve. When a greater number of larvae was crawling or occasionally even swimming, while a considerable number of them was lying quietly, the respiration intensities found were higher (2-3-4). High respiratory rates (9-10) were found in experiments on newly metamorphosed, vividly ventilating animals which attach themselves to the glass by means of their byssus secretions, their shells widely open. High values of respiration/N were also found when all larvae were active at the same time, either swimming, crawling or ventilating in the diver (5-6).

Unfortunately, it proved unfeasible to perform an experiment in the μ l diver in which all or even a reasonable percentage ($> 25\%$) of the larvae were swimming simultaneously.

Accordingly, not before an experiment was made on one larva in a $0.1\ \mu$ l diver, the author succeeded in measuring the metabolic rate of the swimming larva. In three experiments (out of c. 10) a single *Mytilus* larva was swimming vividly in the diver and the metabolism of swimming could be measured. However, the N content of the larva was so low that it could not be determined directly,

each analysis requiring some (6–20) larvae. Fig. 27 shows the relation between the length of the larvae and their N-content. Using this curve and knowing the length of the larva under investigation, the N-content of this larva may be determined with reasonable accuracy. Thus, the respiration/N of the single larva could be

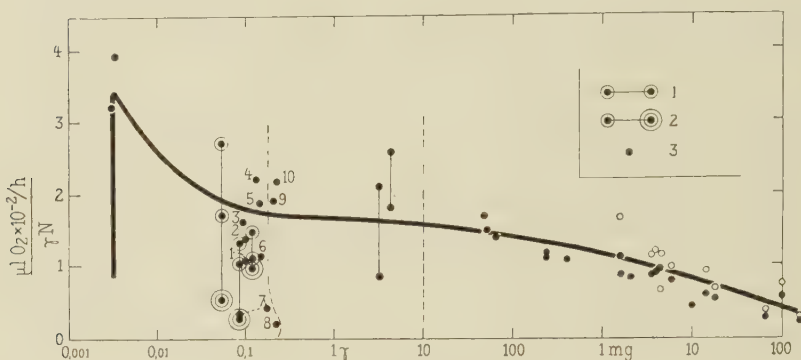


Fig. 28. *Mytilus edulis*.

Relation between respiration/N (ordinate) and body size, expressed by the N-content (abscissa). The experiments to the left of the two dotted lines are performed with larvae from the plankton; the two experiments on the smallest animals refer to 24 hours old trochophorae still without shells. The experiments between the two dotted lines are performed on young bottom stages, < 1.5 mm long. The experiments to the right of both dotted lines are carried out on bottom animals which have been so large that the tissues could be isolated from the shells. N < 10 γ: diver experiments. N > 10 γ: Winkler experiments. Open circles: Respiration/N in the tissue. Full circles: Respiration/total N.

Numbers in the corner:

1. 0.1 μl divers, highest and lowest respiration/N during swimming.
2. 0.1 μl divers, highest and lowest respiration/N during non-swimming.
3. Experiments on a number of animals simultaneously (with the exception of some of the largest animals).

Numbers in the figure proper: cf. the text.

measured in varying states of activity. These determinations are given in Fig. 28 as the points surrounded by circles. Three points which are connected by thin, vertical lines describe each of the three experiments mentioned. The highest point indicates the highest, and the middle the lowest respiratory rate found during swimming, while the lowest point shows the lowest respiratory rate found during rest of the same animal.

Fig. 28 reveals clearly that we cannot possibly distinguish between the metabolic rate of 1) swimming veliger larvae, of 2) veliger larvae which, using the byssus, attach themselves to the support, ventilating vividly and, finally, of 3) the metamorphosed ventilating larvae. Finally, it may be added (cf. Fig. 6, section 3.112) that probably the metabolic rate of the veliger larvae which crawl on the foot is of the same order as that of the animals in the above states of activity. On the other hand, both Figs. 5 and 28 show that the metabolic rate of the resting veliger larva (i. e. the larva lying on the bottom of the diver with its shells closed or only occasionally open) is lower than the metabolic rate of the swimming, crawling, or settled, but vividly ventilating larva.

The above description comprises experiments on trochophorae, veliger larvae and young, metamorphosed *Mytilus edulis* ($N < 10 \gamma$). Except for the trochophorae which have no shells, all nitrogen determinations represent the N-content of the shells plus tissues.

The experiments on the right side of Fig. 28 ($N > 10 \gamma$) are Winkler experiments performed on larger bottom animals. The solid points indicate the metabolism calculated per total N, the circles, however, the metabolism per N of the tissue, only. The curve describes the metabolic rate per total N of all animals. The curve for metabolism per N of the tissues would lie on a somewhat higher level than that drawn in the figure; to the right, its position would be indicated by the circles, to the left its level would be c. 10 per cent higher than that of the curve drawn, because c. 10 % of the total N of veliger larvae is situated in the shells (cf. Fig. 27).

The curve of Fig. 28 represents the author's interpretation of the experimental results. When a curve has altogether been drawn, it is only in order to make it possible in a later chapter of this treatise to compare the experiments of Fig. 28 with those of other graphs published in this paper. The approximate correctness of the curve, however, can hardly be contested.

4.25. Crustacea.

4.251. *Experiments with Winkler Titrations*
(in the period $13/5-3/7$, 1946).

Fig. 29 shows the results obtained from experiments with Krogh syringes. The usual technique (as described in 2.14) was applied; only the experiments on copepods (on the left of the figure) need a special discussion. For some days, the plankton was an almost

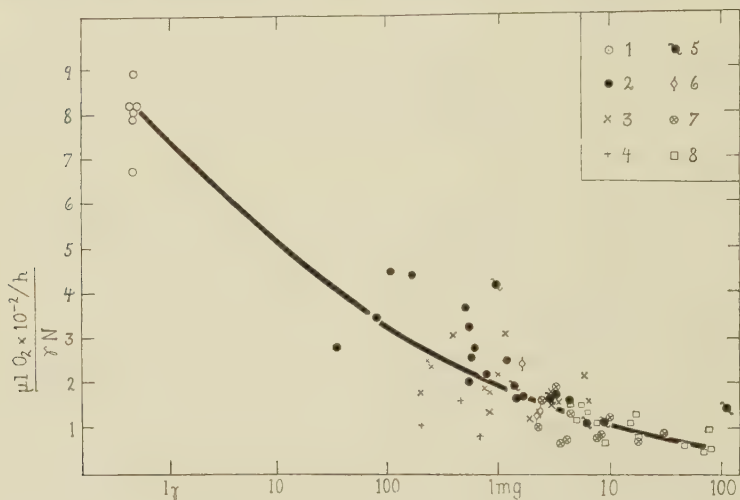


Fig. 29. Crustacea, Winkler experiments.

1. Copepods.
2. *Gammarus*.
3. *Idothea*.
4. *Balanus*.
5. *Carcinus*.
6. *Mysis*.
7. *Crangon*.
8. *Palaemon*.

pure copepod plankton, a fact which enabled us to perform respiration experiments with many copepods in 25 ml flasks. The sample rich in animals was kept in a large Petri dish for one hour. During this period, the animals moved towards the light or the dark side, where they could easily be collected by means of a pipette; subsequently, they were transferred to a filter (of gauze) and washed. From the filter the animals were finally transferred to the flask in which the

respiration experiment should be carried out and which had been filled in advance with water of known oxygen content. After the lapse of 2–3 hours a water sample was carefully drawn for oxygen titration. Since the flask only contained c. 500 animals, it appeared possible to draw the sample without carrying the animals along. Now, the animals were collected on an N-free filter and the animals together with the filter—in the blanks, empty filters—were digested. The experiments showed sufficient agreement. In one experiment negatively phototropic, in five experiments positively phototropic copepods were used. In each case, however, a mixture of species was present, yet the prepondering species was red in the photo-negative sample and bluish in the photopositive samples. The determination of the N-content of the single animal is based on an estimate of the number of animals present in every single experiment. The result (0.5 γ) is in satisfactory agreement with that obtained through direct analysis on equally large individuals (Fig. 30). The negatively phototropic plankton sample showed the lowest respiration intensity, but the difference between this sample and the “positive samples” is only slight and hardly significant.

A closer consideration of the shape of the curve in Fig. 29 reveals a similar decrease with increasing body size of the animals as found in experiments on other groups of animals. The left part of the curve is, however, insufficiently substantiated, but this part is covered by means of the experiments in Fig. 30.

4.252. *Diver Experiments*

(in the period $20/6$ – $25/8$, 1944).

These experiments were performed as discussed in the technical introduction. Some experiments were carried out at the same time on larger animals (the same species as applied in Fig. 29), using the Winkler method. The dotted part of the curve in Fig. 30 is the mean curve for these experiments which, as a whole, were unsatisfactory in technical respect, embracing, however, many useful experiments. Here, the dotted part of the curve is included only in order to show that the curve representing the macro-experiments seems to form a continuation of the curve describing the micro-experiments alone.

The fact that Figs. 29 and 30 are not combined in one figure is due, of course, to the difficulty of doing it easily, the average curve for the experiments from 1944 apparently lying c. 40 %

lower than the average curve for the 1946 experiments. This may be explained in different ways, the most probable explanation being that in 1944 the experiments have been performed somewhat later

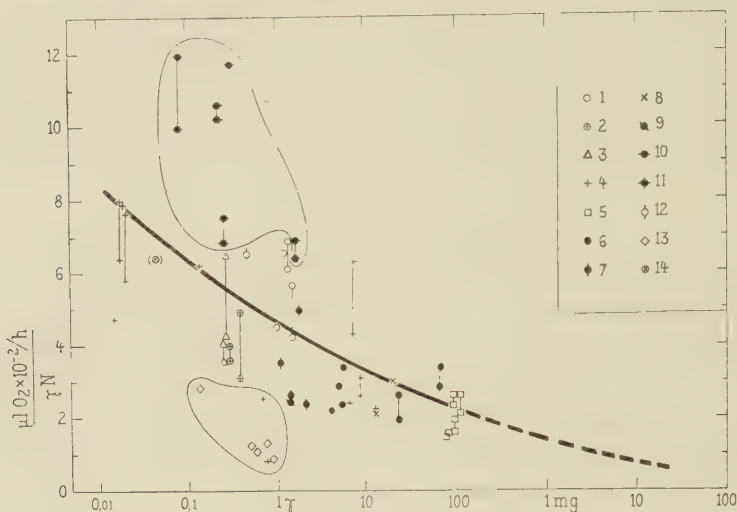


Fig. 30. Crustacea, diver experiments.

1. *Centropages hamatus* (copepod), from plankton.
2. Unidentified copepod, red striped, from sand bottom (2 m), off Snekkersten.
3. *Harpacticid* (copepod) from sand bottom, off Snekkersten.
4. *Balanus* (Nauplius, Cypris-stage and adult (cf. also Fig. 31)).
5. *Palaemon*.
6. *Gammarus*.
7. Isopod from stones off Helsingør.
8. *Idothea*.
9. Crab larva.
10. *Podon*.
11. *Evadne*.
12. Unidentified copepod from plankton.
13. Ostracods from bottom (sand bottom, 2 m, and soft bottom, 20 m).
14. *Chironomus* larva (insect), uncertain determination.

Cladocera are encircled (above), and so are ostracods and cypris-stages of *Balanus* (below).

in the summer than the 1946 experiments. In 1944, the temperature in the Sound was 16–21° C, while it was 12–19° C in 1946, thus on the average 3–4° lower. In recent years, Fox and coworkers (27, 28, 29, 117) have shown that the metabolism and the activity of numerous

animals, measured at a given temperature, depend upon the temperature at which the animals had lived prior to the experiment, the metabolic rate being the higher, the lower the temperature at which the animals had lived previously. These temperature conditions must possibly be taken into consideration in the evaluation of all experiments reported in the present work. It is, however, worth mentioning that in other groups of animals only slight differences (0–20 %) are found between the average of the macro-experiments from 1946 and the average of the preliminary macro-experiments from 1944. If a difference is altogether noticeable, higher values were obtained in the experiments from 1946. The slight difference in season seems thus to be detectable in the whole material; it is, however, hardly significant and has only been taken into consideration in the case of the crustaceans (cf. the discussion).

Figs. 29 and 30 show a marked scattering of the experimental results, especially from species to species. Factors such as living habit, rate of growth, etc. may contribute to this scattering. In the following, some of these factors will be considered separately.

4.253. *Morphology.*

In this connection, the only conspicuous feature was the low respiration of ostracods from the bottom as well as of planktonic cypris-stages of *Balanus* (Fig. 30). The designation cypris-stage indicates the similarity of this animal to the ostracod *Cypris*. The two shells give both types of animals a great, though superficial similarity to a bivalve. The similarity in respiration/N of the two animals may be due to two factors, both of them to be ascribed to their peculiar morphology. Firstly, it is possible that the shells, as compared with the skeleton of other crustaceans, contain a considerable amount of non-respiring N and, secondly, both animals are liable to close their shells for certain periods; thereby, the metabolism may become anaerobic, as is the case with bivalves. Finally, a third, yet less probable possibility exists, *viz.*, that the active tissue proper both in the cypris-stages of *Balanus* and in the ostracod respire very faintly. The author cannot decide which possibility should be preferred, but he is in favour of the assumption that the large shell may contain considerable amounts of N.

4.254. *Activity.*

The influence of the activity upon the metabolic rate of the single animal is not very great, as it results from a previous chapter and as it also appears from Fig. 30. When two points in this figure are depicted one below the other and are connected with a line, these values denote the variation in the metabolic intensity found in the respective experiment. The metabolic rate of the animal at complete rest appears to be c. 50 % of the metabolic rate of the

"normally active" animal (Fig. 12 in 3.113). If it is, moreover, kept in mind that during the experiments the crustaceans (with some few exceptions) most frequently maintained what should be regarded as the "normal activity" of the species, it is clear that a marked scattering of the values ($> 25-50\%$) obtained in different experiments on the same species can hardly be explained as being exclusively due to differences in activity of the animals. Other factors must also play a rôle. In this connection, it should be pointed out that animals of the same species may contain rather varying amounts of N in the intestine. This may be the cause of some deviations in the determinations of the respiratory rates.

When comparing animals of different species, though with the same N-content, the most active animal seems to show the highest metabolism/N. Thus, if we compare the *Cladocera* (*Podon* and *Evadne*) with the copepods *Centropages hamatus* from the plankton or *Harpacticid* and "unknown" from the sand bottom, the activity of the animals can be estimated to decrease in the mentioned sequence, and the same holds for the metabolic rates, as it appears from Fig. 30. Obviously, however, the activity is only one out of many factors determining the metabolism of the single animal and this becomes still more indubitable if we include in our comparison the very vivid nauplii of *Balanus*. These latter animals must be considered just as active as the *Cladocera* but, nevertheless, they have a somewhat lower metabolic rate.

4.255. *Ontogenetic State of Development.*

Considering equally large animals, Figs. 29 and 30 do not indicate any definite difference between the metabolic rate of adult (*Copepods* and *Cladocera*) and growing forms (nauplii of *Balanus*). The present material is very useful in this respect since, no doubt, the growing forms considered are just as free of yolk as are the adults; thus, there is no reason to assume that "passive yolk N" should be included in our N-determinations.

4.256. *Living Habit.*

It seems to be of minor importance for the metabolic rate whether the animal lives on the bottom or it is floating freely in the plankton, whether it is in growth or it is adult. This involves the assumption that the "work of swimming" is small and exceeds neither the "work of creeping on the bottom" nor the "work of growth" (cf. also 3.14, where we arrived at the same conclusion by way of different arguments and for other animals).

4.257. *Species, Group.*

The ostracods were found to show the lowest respiration, the copepods and the nauplii of *Balanus* respire more intensely, and the *Cladocera*, finally, have the highest respiration. These differences may perhaps be ascribed to differences in visible activity of the animals. More cautiously, however, we may just speak of group differences.

Summing up what has been said above, we may state that for comparable animals the body size seems to be predominant in deter-

mining the respiration intensity of the animals. Provided the body size varies sufficiently, the effect of this factor cannot be hidden, yet be somewhat blurred out due to the effect of other factors, *viz.* activity, living habit, etc.

After the above account, the curve-drawing in Fig. 30 becomes somewhat more justified than it would appear incipiently: no consideration has been paid to the ostracods whose morphology, and possibly also physiology (possibility for anaerobiosis), deviate markedly. It may be emphasized again that not too great stress should be laid on the curves of Figs. 29 and 30 which should mainly serve as a synoptical basis for the comparison with other groups of animals. They are published here as the present author's summarizing, subjective interpretation of the experimental results and they are considered the approximate average expression for the relation between metabolic rate and body size (N-content) of the crustaceans in "normal" activity.

4.258. *Balanus*.

Only for one single genus, *Balanus* (*balanoides*(?)) the author succeeded in comparing the metabolism of the larva (nauplius,

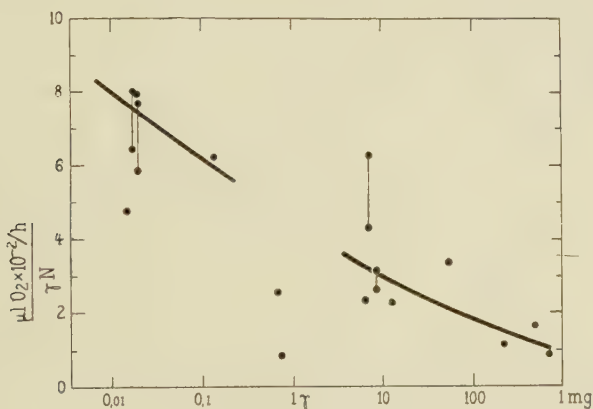


Fig. 31. *Balanus*.

Relation between metabolic rate (ordinate) and body size, expressed by the N-content (abscissa).

Left part of the curve: experiments with nauplii; right part of the curve: experiments with sessile animals. The two experiments on animals the N-content of which is somewhat less than 1 γ are performed with cypris-stages. Since the cypris-stages seem to deviate essentially in respiration intensity from nauplii and adults, the curve has not been drawn through here.

cypris-stage) with that of the adult animal. The result appears from Fig. 31. The interdependence between the body size of the animals and their metabolic rate follows approximately the average curve found for all crustaceans studied (Figs. 29 and 30) and, thus, it has almost the same slope as found for other organisms. The uncertainty with respect to the interpretation of the cypris stage experiments has already been discussed. In view of this uncertainty, the curve has not been drawn through that part of the figure where the cypris experiments are situated. (It should be emphasized that, in this figure, micro-experiments from 1944 are compared with macro-experiments from 1946).

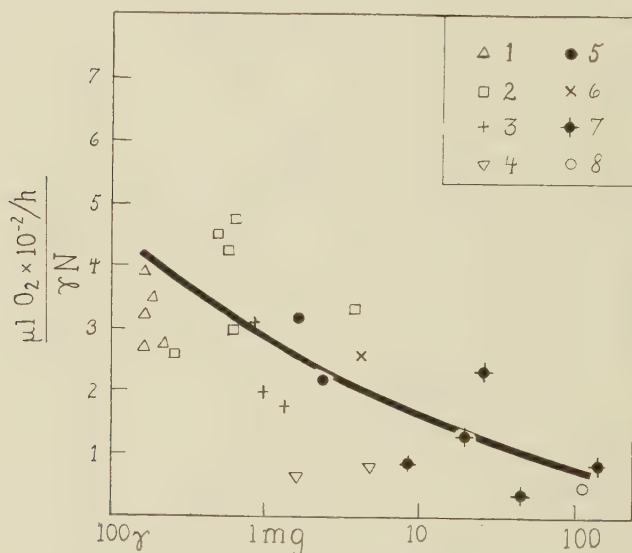


Fig. 32. *Pisces*.

Relation between metabolic rate (ordinate) and body size, expressed by the N-content (abscissa).

1. Spawn, probably belonging to the *Clupeini*.
2. *Belone*, spawn.
3. *Pleuronectidae*, spawn.
4. Unidentified, eel-like.
5. *Syngnathid* fish, sp. I.
6. *Gadus*, spawn.
7. *Syngnathid* fish, sp. II.
8. *Spinachia*.

4.26. Pisces.

Some experiments (Winkler experiments), the results of which are shown in Fig. 32, were performed with different fishes, all of them occasionally caught, and of moderate or small size.

The oxygen consumption as expressed per N unit exhibits a marked decrease towards the right side of Fig. 32 but, perhaps, the curve depicted cannot be regarded as a true size-metabolic rate relation, since all the smallest fishes are planktonic spawn which swim more vividly than do the somewhat larger fishes which were full-grown fishes from the fucus belt (cf., however, what has been said concerning the crustaceans). However, it should be mentioned that the plaice spawn, especially the two lowest respiring individuals, were considerably less active than the other fish spawns.

4.27. Conclusion.

The metabolic rate (metabolism/N) of all the animals considered decreases systematically with their increasing body size. This involves that the metabolism/N of the micro-fauna is higher than the metabolism/N of the macro-fauna. For the same reason, the larval stages of different bottom animals show a higher metabolic rate than the adult bottom animals, but the sedimentation process proper affects only slightly, or possibly not at all, the metabolic rate of the animals studied in the present work. The ratio between the metabolism/N of the micro- and the macro-animals studied is somewhat varying, but, on an average, it can be stated to be c. 4–5 for a variation in body size of 10^4 – 10^5 times.

4.3. *Metabolism/Weight in Relation to the Body Weight of the Animals.*

In the preceding sections, the metabolism of a number of small and medium sized animals from Øresund has been compared on the basis of N-determinations on the bodies. The literature comprises a great number of investigations which, within certain groups of animals, aim at finding a relation between the metabolic rate and the body size, the latter generally defined by the fresh weight of the animals. These studies cannot directly be compared with the author's

own investigations and they will, therefore, be accounted for in a separate chapter. At the same time, the present author's experiments concerning the metabolism/weight will be described.

In the literature available (except, for instance, KROGH (60) and BENEDICT (5)), a clear distinction has not always been made between experiments performed on small and on large forms of the same species (intraspecific comparison) or on small and large individuals of different species (interspecific comparison). Therefore, such a distinction cannot either be made here.

No attempt has been made to quote all papers dealing with the problem of metabolism and body size. We must confine ourselves to mentioning most of those papers which, according to the present author's view, form a valuable contribution to the understanding of the problem and which, moreover, have been accessible in this country. Different investigators measured the metabolic rate of the animals under widely varying conditions. Some workers attempted to work under standard conditions while others, among which the present author, considered it difficult or almost impossible in a comparative investigation to define the concept "standard conditions" and, for this reason, aim at the maintenance of "natural activity"—in so far as this state can be defined properly. The author's experiments were carried out at 16° C, those described in the literature were performed at different temperatures around room temperature (cf. the texts to the figures). From MONTUORI's great work from 1913 (83), dealing with the metabolic rate of a great many marine animals, it would appear that a general tendency towards a decrease in metabolic rate with increasing body size of the animals does not exist. However, a closer examination of MONTUORI's table indicates that this table must contain rather big miscalculations. A conversion by means of a simple division of one column of the table to another column reveals essential errors in 15–20 % of all cases. Since several mathematical manipulations must have been involved in his calculation of the experimental results, it seems rather probable that the miscalculations make MONTUORI's material useless. After MONTUORI's work thus has been discarded, the remaining experiments published in the literature generally indicate a decrease in metabolism/weight with increasing body size of the animals. This will be demonstrated in the following.

4.31. Echinodermata.

HELGA MEYER (81) has performed experiments on individuals of different size of *Asterias rubens*; her results are shown in Fig. 33. The steep decrease in metabolic rate with increasing size of the animals is due probably in part to the fact observed by MEYER herself that the respiration of the adult *Asterias*, owing to a weakly

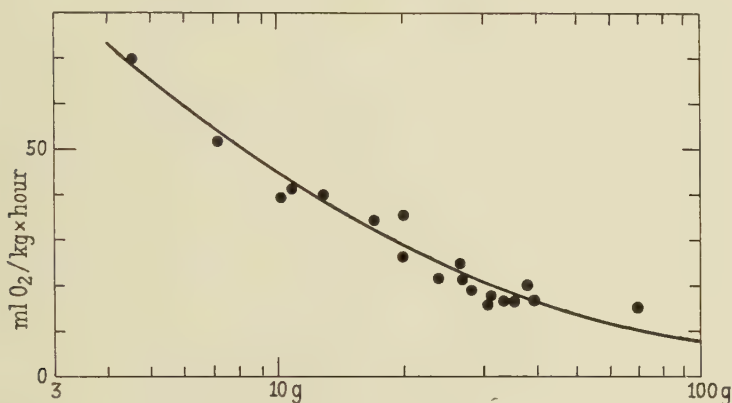


Fig. 33. *Asterias rubens*.

Relation between metabolic rate (ordinate) and body weight (abscissa).

No temperature given, probably 20° C. Recalculated from MEYER's Table 1. (81).

developed circulation, is greatly dependent on the oxygen pressure. This holds also for many other larger species among the lower animals such as, for example, *Actinia* (HENZE (39)): The larger the animal, the smaller a fraction of the body can be supplied with oxygen by diffusion from the surface. On *Holothuria*, COHNHEIM (24) found decreasing metabolic rate with increasing body size of the animal.

4.32. Vermes.

On a large number of marine polychaetes, BOUNHIOL (12) found the CO₂-production/weight to decrease markedly with increasing body size of the animals. The results may be correct, but the technique employed was rather unsatisfactory, because the high solubility of carbon dioxide in water may have led to various errors; therefore, the experiments will not be discussed in greater detail at this place.

On *Planaria agilis*, ALLEN (1) observed a markedly decreasing metabolic rate with increasing body size. The animals were examined after a long period of starvation. Fig. 34 comprises a number of

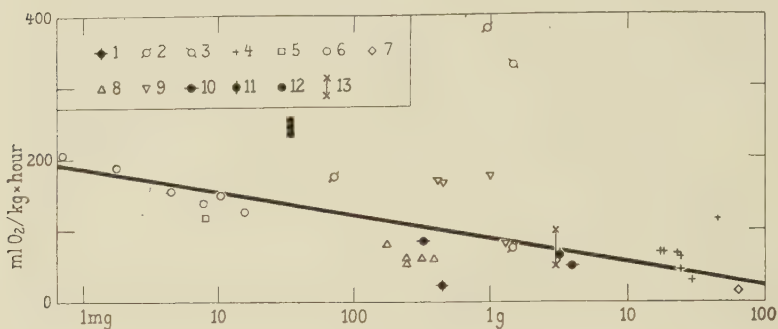


Fig. 34. *Vermes*.

Relation between metabolic rate (ordinate) and body weight (abscissa).

1. *Hirudo*, starving, calculated from PÜTTER (94).
2. *Glycera*, vividly swimming, 13-17° C (COHNHEIM (24)).
3. *Nephtys*, vividly swimming, 13-17° C (COHNHEIM (24)).
4. *Sipunculus*, moving occasionally, only animal with the highest metabolic rate very vivid, 13-17° C (COHNHEIM (24)).
5. *Planaria agilis*, more than 10 days starvation, 20° C, calculated from Fig. 2 in LUND (79).
6. *Planaria agilis*, 22 days starvation, 19.6° C, calculated from ALLEN's (1) Table 10.
7. *Urechis caupo*, 15.4—18.8° C. Average, calculated from HALL's (36) paper.
8. *Lumbricus*, 17.1—19.5° C, LESSER, quoted from KROGH (60).
9. *Lumbricus*, 19.2—20.5° C, THUNBERG, quoted from KROGH (60).
10. *Lumbricus*, average of JOEL's experiments.
11. *Lumbricus*, 20—23° C, KONOPACKI (54), quoted from KROGH (60).
12. *Tubifex tubifex*, 18—20° C, weight of the single animal estimated to be 20 mg, in order to fix the abscissa value of the measurements. From HARNISCH (37).
13. *Nereis virens*, 17—20° C, uppermost point: metabolic rate in normal activity, lowest point: metabolic rate in narcotized animals. Mean values. LINDROTH (76).

experiments from the literature among which we find ALLEN's above mentioned ones, but not those of BOUNHIOL. The experimental conditions (temperature, activity, etc.) are described in the text to the figure. The temperature varies in the various experiments between c. 15° C and 23° C. The two highest points refer to COHNHEIM's (24)

experiments from 1912 on swimming *Glycera* and *Nephtys*. All other points refer to experiments on animals which in different states of activity lie on the bottom of the experimental vessel. LUND's (79) and ALLEN's (1) experiments on *Planaria agilis* are the only experiments which have been carried out after a long period of starvation; presumably these results, therefore, are relatively low. The body size of the animals dealt with in Fig. 34 varies by a factor of c. 10^5 . The figure exhibits a rather definite, but not very marked decrease in metabolic rate with increasing body size.

In experiments on *Ascaris lumbricoides* (weight 0.3–10 g) performed at 37° C, KRÜGER (65) found the respiratory rate to decrease markedly with increasing body size of the animals, but approximate proportionality seems to prevail between metabolism and surface; this proportionality, however, KRÜGER regards only as incidental. KRÜGER's experiments seem to disagree with the general impression conveyed by Fig. 34 (cf., however, the discussion, Section 5.23).

4.33. Mollusca.

Fig. 35 shows the author's own experiments on the bivalve *Mytilus edulis*; Fig. 36 illustrates experiments on the prosobranch

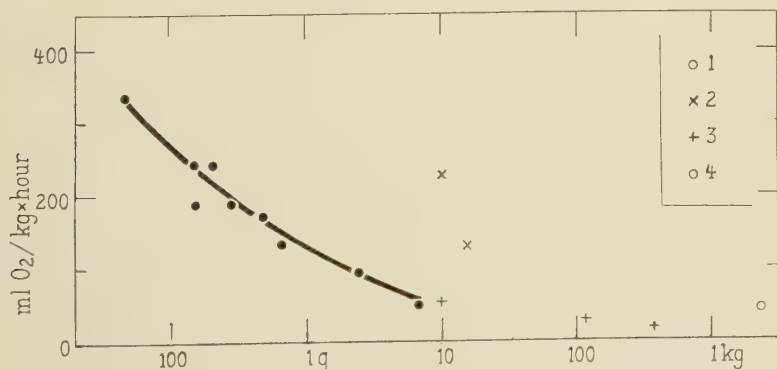


Fig. 35. *Mytilus edulis*.

Relation between metabolic rate (ordinate) and weight of tissues (abscissa). The author's experiments. Some determinations (from the literature) on Cephalopods are included in the figure.

1. *Mytilus edulis*, 16° C, the author's experiments.

2. *Eledone moschata*, 15° C, COHNHEIM (24).

3. *Octopus vulgaris*, 13.3° C, BUYTENDIJK (21, 22).

4. *Octopus vulgaris*, 14–19° C, JOLYET and REGNARD (46).

All animals 2, 3, 4 are almost at rest.

snails *Nassa reticulata* and *Littorina littorea*. In both instances, the decrease in metabolic intensity with increasing body size of the animals is apparent. WEINLAND (112) stated that *Anodonta* obey the surface law; this statement, however, cannot be maintained with certainty, because the size of the animals varies by a factor 3, only. In an interspecific comparison of different species belonging to the

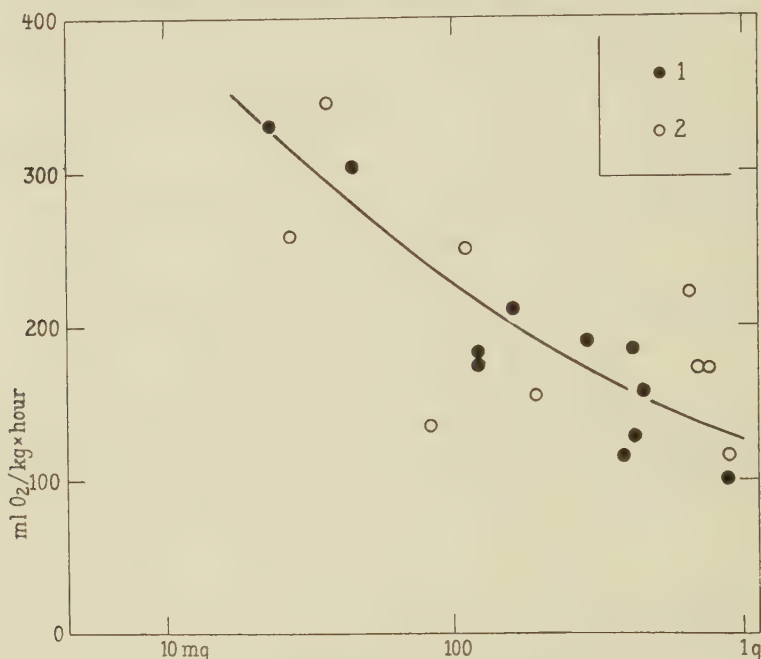


Fig. 36. *Nassa reticulata* (1) and *Littorina littorea* (2).

Relation between metabolic rate (ordinate) and body weight of the tissues (abscissa), the author's own experiments. 16° C.

genus *Helix*, LIEBSCH (71) found the metabolism to vary linearly with the body weight, *i. e.* the metabolic rate does not depend on the weight of the animal.

With regard to the cephalopods only scattered informations can be found in the literature. BUYTENDIJK's (21) and COHNHEIM's (24) results on resting *Octopus vulgaris* and on resting *Eledone moschata*, respectively, comprised in Fig. 35, should be compared with the curves for *Mytilus* and gastropods (Figs. 35 and 36). Probably, the metabolism of cephalopods is higher than that of equally large lower molluscs, *i. e.* gastropods and bivalves, but the material forming the basis of this conclusion is only very scanty.

4.34. Arthropoda.

Fig. 37 represents the author's own experiments on various crustaceans. The decrease in oxygen consumption/weight with

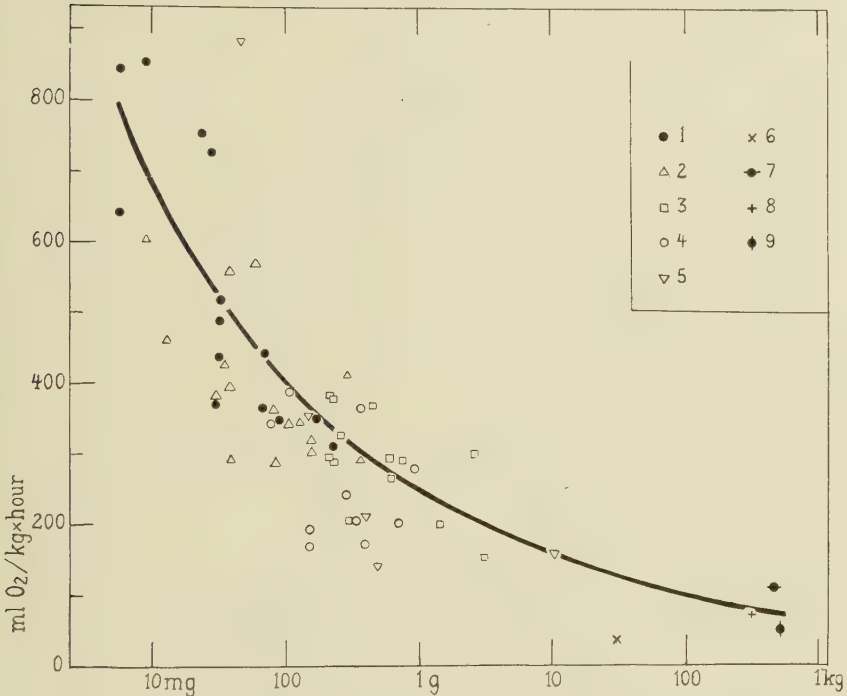


Fig. 37. Crustacea.

Relation between metabolic rate (ordinate) and body weight (abscissa).

- | | | |
|----------------------------------|---------|--------------------------------|
| 1. <i>Gammarus</i> | 16° C | } The author's own experiments |
| 2. <i>Idothea</i> | 16° C | |
| 3. <i>Palaemon</i> | 16° C | |
| 4. <i>Crangon</i> | 16° C | |
| 5. <i>Carcinus</i> | 16° C | |
| 6. <i>Astacus fluviatilis</i> | 12.5° C | } JOLYET and REGNARD (46) |
| 7. <i>Cancer pagurus</i> | 16° C | |
| 8. <i>Homarus vulgaris</i> | 15° C | |
| 9. <i>Palinurus quadricornus</i> | 15° C | |

increasing body size of the animals is obvious. Some few measurements by JOLYET and REGNARD (46) on large decapods are included in the figure.

KITTEL (49) has promised to publish her experiments on cru-

staceans. They would be of great interest because this investigator has earlier obtained very deviating results on insects (cf. below).

WEYMOUTH, CRISMON, HALL, BELDING and FIELD II (114) performed experiments at 15° C on the crab *Pugettia producta* varying in size from 1–300 g. Within the range considered, their results are c. 30 % below the average curve of Fig. 37. These experiments first came to the present author's knowledge after Fig. 37 had been drawn and, therefore, they are not included in this figure. The agreement between Fig. 37 and the experiments mentioned above can, however, be considered satisfactory. Experiments on slices of the hepatopancreas from large and small crabs showed the metabolism/weight of this organ to decrease in the same way with the body size of the animals as does the total metabolism of the animal.

A comprehensive literature deals with the relationship between metabolic rate and body size of insects. Almost all authors find the lowest metabolic rate in large forms but, on the whole, the results are greatly deviating.

The class of the insects is so well differentiated that each species or small group of animals may often have acquired its own characteristic respiration intensity. This feature is not so pronounced in other groups of animals and, therefore, we can hardly establish a law embracing the metabolic intensity of all insects. Thus, the decrease in metabolic intensity with increasing body size of the animals must be studied for each species separately. It should, however, be mentioned that, if one and the same species has been the object of repeated investigations by different authors, the results will appear to deviate considerably.

In insect experiments, the metabolic rate is almost always expressed as a function of W^e . For a great number of different insects (imagos of *Coleoptera* and *Diptera*, larvae of *Vanessa io* and *urtica*, larvae of *Tenebrio molitor*) KITTEL has found the metabolism to vary with the weight of the animals ($e = 1.00$) rather than with their surface ($e = 0.67$). BORNEBUSCH's (11) results valid for the insects of the forest soil (embracing mostly species belonging to the *Coleoptera*) indicate a low value of e . His results are computed in Fig. 38. From the curve drawn we find e to be equal to c. 0.7. For the grasshopper *Melanoplus*, BODINE (7) found $e =$ c. 0.67 ("surface law"), and the same result was obtained by BUTLER and INNES (20) working on the metabolism of *Locusta*. For *Blatta* species, GUNN (35) found $e = 0.75 - 0.8$, and for *Tenebrio* larvae different authors obtained

largely varying results, frequently not the same value for e in different ranges of body size of the same animal. In MICHAEL's experiments (82) e actually varies between 1 and 0.3. From TEIS-

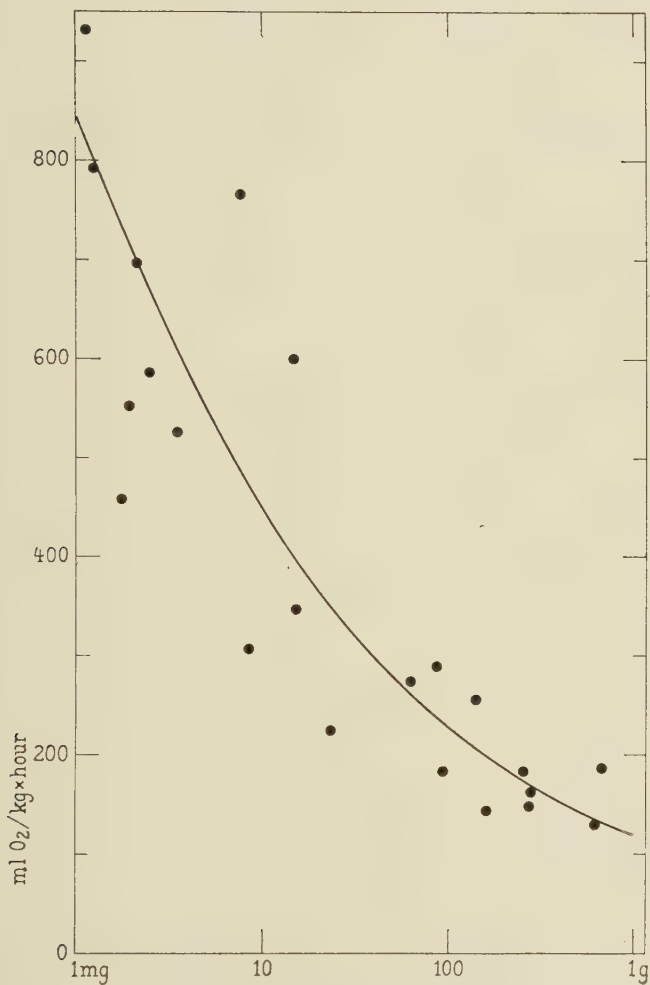


Fig. 38. Insects from forest soil.

The values are calculated from BORNEBUSCH's Table 21 (11), 13° C.

SIER'S (105) experiments on the same animal (misinterpreted by BUDDENBROCK (19)) it seems to result that e is dependent on the diet of the animal and especially on whether the animal is fed or starved (e varies between 1 and 0.7). Fig. 39 shows one of TEISSIER's

experiments with starving larvae (calculated from Table 19 in TEISSIER's work) at 20.4° C.

Thus, with a few important exceptions, agreement seems to prevail with respect to the fact that the metabolic rate of the insects decreases with increasing body size, as it has also been found for other animals; however, the different authors disagree concerning the more accurate value of the functions.

It should be emphasized that in all experiments on adult insects the different authors have aimed at "standard" conditions. This

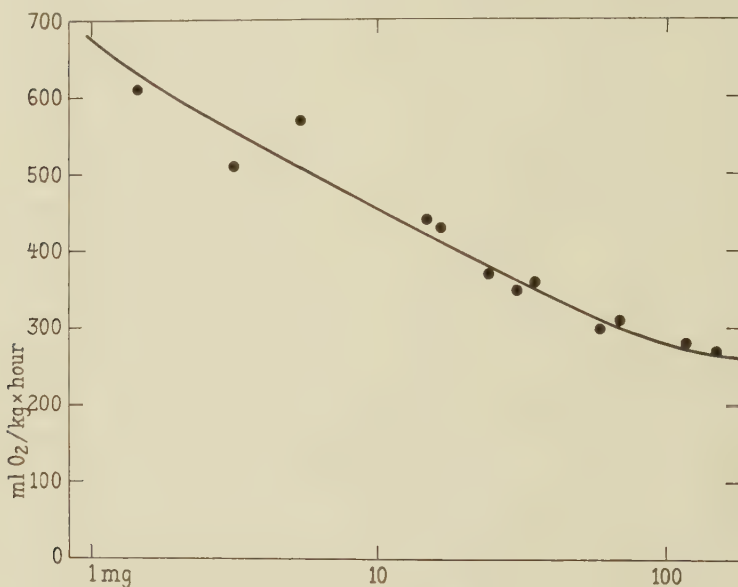


Fig. 39. *Tenebrio molitor*.

Relation between metabolic rate (ordinate) and body weight (abscissa), calculated from TEISSIER's (105) Table 19. 20.4° C.

is easily understood, because an even invisible activity from the side of the animals may seriously affect the result; the energy expenditure may increase to a multiple of the resting metabolism. Standard conditions, however, are very difficult to obtain. The extreme variability of the metabolism of the single insect, therefore, is a serious hindrance for the establishment of exact metabolism-size relationships for insect imagos.

4.35. Pisces.

Generally, the metabolic rate of fishes has been described to decrease with increasing body size of the animals. Some authors (RUBNER (100)) believe in a proportionality between surface and metabolism, other investigators, however, (JOLYET and REGNARD (46), BUYTENDIJK (21), and MORGULIS (84)) do not agree. In Fig. 40, the author's own experiments (at 16° C) with different marine fishes are plotted on one curve¹ together with RAFFY's (95) experiments at 17° C on eels in various stages of development. The metabolic rate of the elver was measured by RAFFY at 13° C; in view of the low temperature, the value obtained may be relatively too low. In this main figure also TERROINE and DELPECH's experiments (106) on large eels are included, although they deviate considerably. As it will be shown, experiments on tenches performed by the same authors do not deviate from the ordinary curve picture. In order to avoid a too dense accumulation of points in the right part of the figure, some experiments are presented separately in the figure inserted in the corner, *viz.* BUYTENDIJK's (21) experiments on marine fishes, JOLYET and REGNARD's (46) old experiments on marine- and fresh water fishes, and KNAUTHE's (53) experiments on carps. This separate figure is drawn in the same coordinate system as the large one and, by means of simple parallel displacement, it can be brought to coincide with the latter.

From the author's own experiments presented in Fig. 40 it seems to result that the metabolic rate decreases markedly with increasing body size of the fishes. However, as mentioned earlier, in these experiments pelagic fish spawn is compared with the full grown fishes of the fucus belt, a comparison which perhaps is inappropriate. The curve describing the author's own experiments may, however, easily be continued by means of RAFFY's results on eels, which also exhibit an unambiguous and marked decrease in metabolic rate with increasing body size of the animal. SPÄRCK's (102) experiments on eel larvae², which seem to indicate that the metabolic intensity increases by a factor ten with increasing body size of the yolk-free larva, and TERROINE and DELPECH's (106)

¹ In the author's own experiments, one and the same animal has often been used in both Fig. 32 and Fig. 40, once applying the body weight and the other time the N-content.

² Not included in Fig. 40.

experiments which show an extraordinarily low metabolic rate for fully developed eels, make RAFFY's results much less certain. The drawn curve agrees, however, rather well with BUYTENDIJK's results,

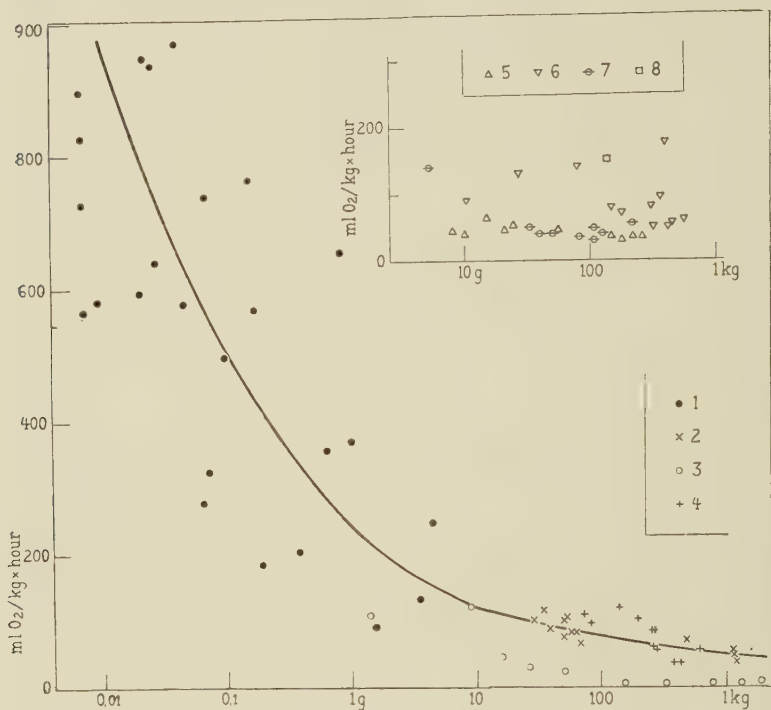


Table 40. *Pisces*.

Relation between metabolic rate (ordinate) and body weight (abscissa).

- Main figure:
1. Own experiments. Generally, the same animals as used for the experiments in Fig. 32. 16° C.
 2. Eels (*Anguilla vulgaris*), 17° C, however 13° C in the experiments on the smallest animals (elvers). RAFFY (95).
 3. Eels, 20° C, TERROINE and DELPECH (106).*)
 4. Tenches (*Tinca vulgaris*), 20° C, TERROINE and DELPECH (106).
- Inserted figure:
5. *Scyllium*, *Scorpaena*, *Crenilabrus*. No temperature given. BUYTENDIJK (21, 22).
 6. Marine fishes, 13-19° C, JOLYET and REGNARD (46).
 7. Fresh water fishes, 11.5-14° C, JOLYET and REGNARD (46).
 8. Carps (*Cyprinus carpio*), 20° C, average of KNAUTHE's experiments (53).

*) Error: the point representing the smallest animals is RAFFY's experiment on elvers.

with JOLYET and REGNARD's experiments on fresh water fishes, and with TERROINE and DELPECH's results on tenches of varying size. These latter groups of experiments all show the metabolic rate of the fishes considered to decrease with increasing body size, and the average metabolic rate agrees well with the curve drawn, especially if the experimental temperatures are taken into consideration. JOLYET and REGNARD's experiments on marine fishes do not show the decrease in metabolism with increasing body size. However, the experimental material is very scanty and seems to be unsuited for further theoretical considerations. The authors themselves emphasize that the experiments indicate a more intense respiration of marine organisms than of limnic ones. This may be so; however, it is worth mentioning that the experiments on the former were carried out at somewhat higher temperatures ($13-19^{\circ}\text{C}$) than those on the latter ($11.5-14^{\circ}\text{C}$). As a whole, the experiments seem to indicate that the decrease in metabolic rate, which appears well established in the left part of Fig. 40, most probably continues also in its right part, but the uncertainty of this finding is so high that it is completely impossible to predict anything concerning the metabolism of large fishes. Measurements in this field would be of the greatest interest.

4.36. Amphibia.

In the literature, a number of experiments are available which aim at relating the metabolic rate with the body size of the *Amphibia*, viz. REGNAULT and REISET (96), JOLYET and REGNARD (46), POTT (92), BUYTENDIJK (22), LIANG, CHUNG MOU (70), KREHL and SOETBEER (57), and GROEBBELS (32, 33). The most important contributions to the problem, however, are those of TERROINE and DELPECH (106) and of BLANK (6), both groups of experiments being comprised in Fig. 41. From the work of the French authors only the experiments on male frogs are included, since the large amounts of eggs developing in the larger females (in June) seem decisively to affect the metabolic rate of these animals; hence, less well-defined interrelationships for size and metabolic rate are obtained on females than on males. BLANK's as well as TERROINE and DELPECH's experiments on male frogs agree in showing a rather steep dependence between body weight and metabolic rate. The variation in body size of the animals amounted, however, only to c. 10 times. GROEBBELS (33), when performing experiments on quite small frogs shortly

after metamorphosis, found a metabolic rate not very different from that of the adult frogs. GROEBBELS' experiments, however, loose some of their value because, earlier (32), the same author had published quite different results, using another technique, GROEBBELS does not attach too much importance to this fact, but in his

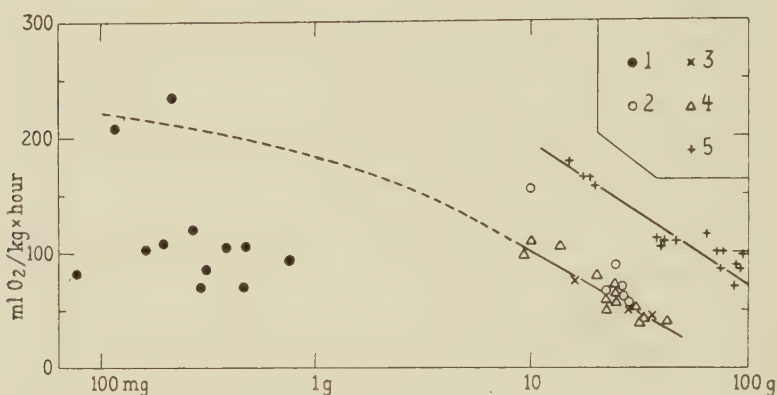


Fig. 41. *Amphibia* (frogs).

Relation between metabolic rate (ordinate) and body weight (abscissa).

1. Young, just metamorphosed frogs, 15.4° C, GROEBBELS (33).
2. *Rana esculenta*, 20° C, TERROINE and DELPECH, Table III (106).
3. *Rana temporaria*, 20° C, TERROINE and DELPECH, Table I (106).
4. *Rana temporaria*, 20° C, TERROINE and DELPECH, Table II (106).
5. Frogs (species not indicated), 26° C, BLANK (6).

last work (using the Warburg technique) he pointed out that the first experiments had yielded false results because of an imperfect technique. GROEBBELS' last experiments included in Fig. 41 show a not insignificant scattering. In evaluating the experiments of Fig. 41 it must be kept in mind that GROEBBELS worked at 15°4, TERROINE and DELPECH at 20°, and BLANK at 26° C, and that no corrections for the temperature were made when inserting the values in Fig. 41. The values are comparable only if we attempt to introduce reasonable corrections for the temperature differences mentioned. The author has made such rough corrections when drawing the curve. However, the dotting to the left is meant to remind the reader of the uncertainty of the curve. After the drawing had been completed, the author through the book of BRACHET (13) became acquainted with the work of WILLS who found the metabolic rate of tadpoles not to be essentially different from that of adults. This seems to agree well with the general feature of Fig. 41.

4.37. Reptilia.

For the reptiles a large series of investigations into the metabolism/weight problem are available. All authors attempted to perform standard metabolism measurements. The most significant and most extensive experiments are those by BENEDICT (3), which are discussed very thoroughly in "Physiology of Large Reptiles". For alle reptiles, with the exception of tortoises, BENEDICT finds that

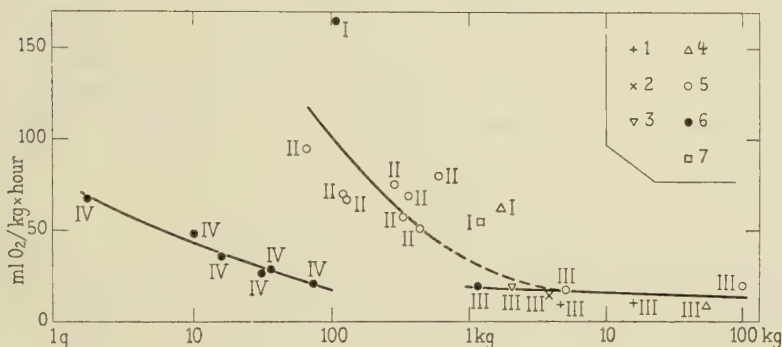


Fig. 42. Reptiles.

Relation between metabolic rate (ordinate) and body weight (abscissa).

- | | |
|--|--------------------------|
| I. KREHL and SOETBEER (57), 25.3° C. | } Basal heat production. |
| II. TERROINE and DELPECH (106), 20° C. | |
| III. BENEDICT (3), 21° C. | |
| IV. KRAMER (55), 20° C. | |
1. *Boa*.
 2. Rattlesnake.
 3. Gopher snake.
 4. Alligator.
 5. Tortoises (minus shells).
 6. Different lizzards.
 7. *Uromastix*.

the metabolism (intraspecifically) is almost proportional to the surface, in tortoises, however, almost proportional to the body weight.

In Fig. 42, we find KREHL and SOETBEER's experiments (at 25.3° C) on *Lacerta*, *Uromastix* and *Alligator* (57) and, moreover, TERROINE and DELPECH's (106) experiments (at 20° C) on different reptiles, the experiments on large reptiles performed by BENEDICT (3) and, finally, KRAMER's night experiments (20° C) on different species of *Lacerta* (55). The subsequent (56) small corrections published by KRAMER have not been taken into consideration. The values de-

scribing BENEDICT's experiments are averages which have been calculated from Tables 58 and 96 and, moreover, from Figs. 48, 63, 65, 83, 88, and 97 of BENEDICT's paper.

The three first mentioned series of experiments were performed after a period of starvation the duration of which varied and, sometimes, was unknown; but the animals were awake and most frequently not quite passive. KRAMER's experiments, however, were carried out during the night on starving, sleeping *Lacerta* of different species and of different sizes. KRAMER finds an exceptionally low metabolism which is very nearly proportional to the surface. For the sake of comparison with experiments on other animals which have been in a state of low or "normal activity" during the measurements, stress should be laid upon the three first mentioned experiments dealing with the metabolism at low activity and during day time. TERROINE and DELPECH's experiments on tortoises indicate only a slightly decreasing metabolism with increasing weight (cf. BENEDICT's above experiments), but for the other, relatively small reptiles the metabolic rate decreases more steeply with increasing body size; for these animals it can well be pointed out—as has, for example, been done by TERROINE and DELPECH—that an approximate proportionality between surface and metabolism exists, if only we are fully aware of the fact that the experiments allow this conclusion to be drawn only in approximation.

With regard to the large reptiles, BENEDICT emphasizes that within the single species the basal metabolism varies more closely with the surface than with the weight. The tortoises form an exception from this rule, the metabolism being almost proportional to the weight. After inserting BENEDICT's average results into Fig. 42 we observe, however, that when comparing these large animals interspecifically, the metabolism per kg of body weight cannot be stated to decrease very markedly.

It would, no doubt, be bold to draw a smooth curve through the heterogeneous experimental material collected from different authors and comprised in Fig. 42. KRAMER's experiments are very instructive in this respect. Therefore, it was considered preferable to draw three separate curves representing the experiments of KRAMER (observations IV), those of TERROINE and DELPECH plus those of KREHL and SOETBEER (observations I and II) and, finally, those of BENEDICT (observation III). The uncertainty, especially of the two latter curves, should, however, be emphasized. The meta-

bolic rate of the snakes seems to be variable, and the tortoises are but remotely related to other now living reptiles (KUHN, 66 and 67). When disregarding KRAMER's experiments as being true only for the state of sleeping, it becomes clear that all other experiments represented in the figure are best described by a bent curve (partly dotted) which, in the right part of the figure, approaches parallelity with the X-axis.

BENEDICT points out that the large tortoises (the largest one investigated by him weighed 169 kg) belong to the largest cold-blooded animals of the present world. Some large fishes and cephalopods, however, reach a weight of up to several 100 kg or even more (*Selache maxima*, *Thynnus vulgaris*, *Architeuthus*), and these forms must be kept in mind in the discussion to come.

4.38. Aves and Mammalia.

RUBNER's previously mentioned experiments which indicate that the metabolism of dogs of most different weights varies with the surface of the animals and not with their body weight, have initiated an intense discussion in the literature. For about 30 years, BENEDICT, the Head of the Nutrition Laboratory in Boston, offered a considerable part of the work capacity of his laboratory to the elucidation of the body size-metabolism relations. He and his collaborators investigated a large number of warm-blooded animals, comparing very small species (mouse and dwarf-mouse) with big ones (bull, steer, elephant). Also the nearest related cold-blooded animals, *viz.* the reptiles, were made the object of closer study. It was aimed at comparing the basal metabolism of all these animals. Hence, care was taken always to measure the metabolism under standard conditions, *viz.* rest, starvation, neutral external temperature, adaptation to the experimental conditions. The studies performed on the reptiles have been discussed above. Therefore, only the measurements on birds and mammals will be dealt with at this place. Before retiring from his position as the head of his laboratory, BENEDICT (1938) has published a valuable and most desirable review of the studies performed in Boston (5). In this review, the comparison of the animals on the basis of the surface unit is secondary to the more direct comparison on the basis of the weight of the animals. In this respect, BENEDICT's book deviates somewhat from earlier publications from the Nutrition Laboratory. This appears natural since the book

definitely abandons the usual concept of the general validity of the surface law for warm-blooded animals.

BENEDICT'S review "Vital Energetics, a Study in Comparative Basal Metabolism", contains diagrams illustrating the way in which the metabolism varies with the body weight within the single species. Generally, the metabolism per kg body weight decreases with increasing weight of the animal. Exceptions may, however, occur (for example, cat and cock). A general surface law, *i. e.* a general

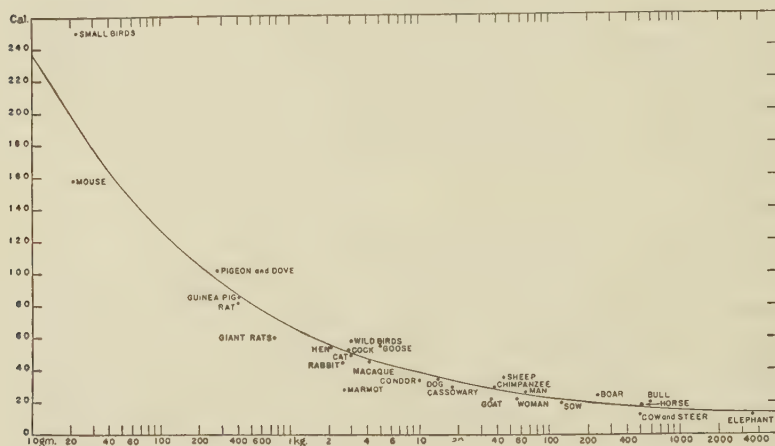


Fig. 43. Birds and mammals.

Relation between basal metabolism (ordinate) and body weight (abscissa).

BENEDICT (5).

proportionality between surface and metabolism, as postulated by RUBNER (98), seems not to be in question, although such a proportionality is frequently more or less approached. It should, however, be mentioned that, in most cases, the body weight of the animals studied varies essentially less than 10 times. Despite all efforts, therefore, accurate relations cannot be established.

In the present connection, however, BENEDICT'S interspecific comparisons are most important. In a summarizing figure (Fig. 43 of the present paper), the metabolic rate of all warm-blooded animals studied is plotted against the body weight of the animals. Each point represents the average of a thorough experimental series. Consequently, the scattering of the points must be considered to give a reliable picture of the ratio between the metabolic rate of different species. At this place, however, we shall not deal with the

possible significance of the fact that one species has a higher metabolism than another equally large species. It might just be mentioned that, generally, the metabolic rate of birds is somewhat higher than that of equally large mammals. BENEDICT explains this as a consequence of the comparatively high body temperature of most birds.

We are mainly interested in the position and in the general shape of the average curve drawn by BENEDICT. This curve was the

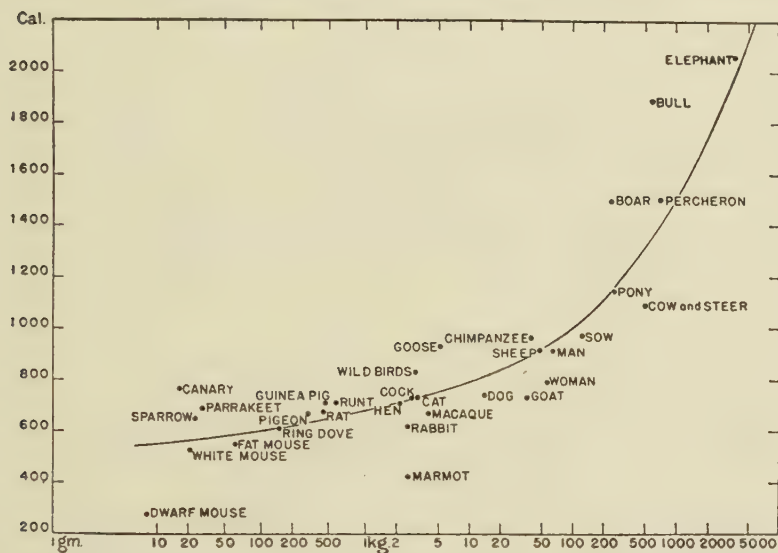


Fig. 44. Relation between the basal heat production calculated per m^2 of different warm-blooded animals and the body weight of the animals. BENEDICT (5).

only one found in the literature which could be directly compared with the author's own curves. On the abscissa (logarithmic scale), the average weight of the animals considered is plotted, and the ordinate shows the metabolic rate in calories per 24 hours per kg.

Hence, the figure illustrates the relation between the basal metabolic rate (in so far as it can actually be defined) and the body weight of the warm-blooded animals. The metabolism measurements, of course, are valid for the normal body temperature of the animals which was measured during the experiments. The shape of the curve will later be discussed in detail. From these and the above mentioned experiments, BENEDICT concludes that neither when compared interspecifically nor when compared intraspecifically do the

warm blooded animals follow RUBNER's surface law. The metabolism, when calculated per surface unit, is not a constant value, but increases steadily with increasing body weight of the animals. This is illustrated by Fig. 44 which is reproduced from BENEDICT's paper. According to BENEDICT (5), KLEIBER (50), and BRODY (14), the basal metabolism of warm blooded animals is approximately proportional to $W^{0.73}$; this is another way of expressing what is depicted in Figs. 43 and 44.

4.39. Conclusion.

From experiments available in the literature on the metabolism/weight of animals it seems to result that the metabolic rate of the animals almost always decreases with increasing body size. In this respect, agreement prevails between the statements to be found in the literature and the author's own experiments on small marine animals (concerning metabolism/N in relation to the body size of the animals).

4.4. *Comparison and Analysis of the Body Size-Metabolic Rate Curves.*

4.4I. Metabolic Intensities in the Animal Kingdom as Compared on the Basis of Metabolism/N and Metabolism/Weight.

The experimental material presented in the preceding sections permits a very rough comparison of the metabolic intensities of different groups of animals. If the N-content of the tissues of a living animal is known (N being expressed as weight % of the wet weight of the animal), metabolism/N values can be converted to metabolism/weight values, and *vice versa*.

4.4II. *Comparison of the Metabolism of Animals on the Basis of the Metabolism/N Value.*

The literature comprises scattered statements concerning the N-content of some animals. Assuming these values to be of general validity also for larger groups of animals, the appertaining curves representing metabolism/weight can be converted into metabolism/N

and can, thus, directly be compared with the present author's metabolism/N curves. This has been done in Fig. 45, where the author's curves are plotted together with curves representing *Amphibia*, *Reptilia*, *Aves*, and *Mammalia*, recalculated from the data of other authors. A considerable number of metabolism/weight curves reproduced in this study were omitted in Fig. 45, however mainly because they would make the figure too difficult to survey. Fig. 45 thus, in the first place, facilitates a comparison of the author's own curves

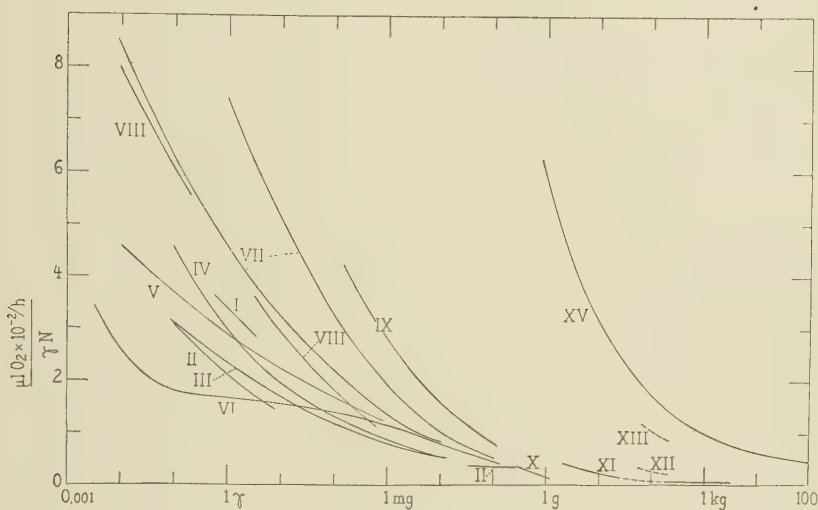


Fig. 45. Comparison of the metabolic rate of the animals on the basis of the body-N. Ordinate: metabolism/N. Abscissa: body-N.

The curves drawn are identical with those published in the present paper, viz.

- | | |
|--|---------------------------------------|
| I. <i>Coelenterata</i> , from Fig. 20, | 16° C. |
| II. <i>Asterias</i> , from Fig. 21, | 16° C. |
| III. <i>Vermes</i> , from Fig. 22, | 16° C. |
| IV. <i>Nassa</i> , from Fig. 25, | 16° C. |
| V. <i>Littorina</i> , from Fig. 26, | 16° C. |
| VI. <i>Mytilus</i> , from Fig. 28, | 16° C. |
| VII. <i>Crustacea</i> , from Figs. 29, 30, | 16° C. |
| VIII. <i>Balanus</i> , from Fig. 31, | 16° C. |
| IX. <i>Pisces</i> , from Fig. 32, | 16° C. |
| X. Frogs, from Fig. 41 (T. and D.), | 20° C. |
| XI. Reptiles, from Fig. 42 (I + II + III), | 20-25° C. |
| XII. Marmot, 16° C. } | calculated from BENEDICT (4) and (5). |
| XIII. Marmot, 37° C. } | |
| XV. Birds and mammals, from Fig. 43, | 37-40° C. |

(I—IX) (left part of the figure). When comparing equally large animals (vertical comparison) it becomes clear 1) that it is impossible with certainty to distinguish between the metabolism/N among the lower groups of animals, viz. *Coelenterata* (I), *Echinodermata* (represented by *Asterias* (II), *Vermes* (III), and *Mollusca* (IV, V, and VI)¹. This is quite clear in view of the scattering of the points around the given average curves. 2) It is of interest that the metabolism/N of the very watery *Coelenterata* (I) cannot with certainty be distinguished from the metabolism/N of the other groups mentioned previously (II—V (perhaps VI included)). Had the comparison been based upon the metabolism/weight unit, however, we should have found the jelly fishes to respire exceptionally faintly. The nitrogen of these animals, therefore, is “just as active” as that of other animals, it is only somewhat more dilute with water. Before final conclusions can be drawn on this point, however, we need separate determinations of respiration/N of the jelly and of the marginal sense organs, respectively. It is very possible that the main part of both respiration and N is localized in the organs, such as the marginal sense organs and the muscles, showing perhaps a normal N-content. 3) There is some indication of a systematically increasing respiration/N when we go from the mentioned lower groups of animals to the crustaceans (VII and VIII). In this connection, it is of interest and maybe also of some importance that in the animal kingdom cross-striated musculature appears for the first time among the *Arthropoda*. It must, however, be emphasized that the increased respiration/N of the crustaceans as compared with that of the lower invertebrates is most easily detectable for the micro-fauna. 4) The metabolism/N of the fishes (IX) is higher than the metabolism/N of all equally large invertebrates studied. The curves now following (X—XV) are recalculated from values found in the literature² and they are included here mainly in order to demonstrate that the whole picture of the curves of Fig. 45 is not essentially different from that of Fig. 46 representing the metabolism/weight of the animals. The

¹ The *Mytilus* curve (VI) alone is hardly sufficient to influence this conclusion, *i. a.* because we do not know whether this curve is typical of the molluscs as a group.

² N % used, 2.2 % in <i>Amphibia</i>	}	mean of the values given by LIANG (70)
2.9 % in <i>Reptilia</i>		
2.5 % in <i>Mammalia</i>		mean of the values given in Tb. Biol.
2.5 % in <i>Aves</i>		estimated as <i>Mammalia</i> .

further comparison of metabolic rates is, therefore, most easily performed on the basis of Fig. 46, where the material available in the literature is presented without conversion to metabolism/N values.

4.412. *Comparison of the Metabolism of Animals on the Basis of the Metabolism/Weight Value.*

In Fig. 46 the author's metabolism/N measurements are converted into metabolism/weight on the assumption that the N-percentages found for the adult marine animals are valid also for the larvae of the same animals or for other micro-animals belonging to the same zoological group¹. In this way, the curves I—VI were obtained. According to NEEDHAM (85), however, small animals usually contain more water than larger ones. This could not be taken into consideration in the present treatise. Possibly, therefore, the respiration intensity of very small animals, when calculated per wet weight, is actually lower than depicted in Fig. 46. It is more important, however, that Fig. 46 represents the respiration intensity of animals of almost the same percentage water content. The *Asterias* curve of Fig. 21 has not been converted in view of the especially low N % of this animal (0.9—1 %), and the same is true for the *Coelenterata* (Fig. 20) which contain very much water. From the curves discussed in 4.3 only two have not been included here, *viz.* the *Tenebrio* curve (Fig. 39) due to lack of space, and the *Lacerta* curve (Fig. 42, IV), the only curve valid for a sleeping animal. The curves VII—XVI represent experiments performed at 16° C (own experiments and a few others) or at temperatures around 16° C (cf. the text to the figure). No attempt has been made to correct the curves for deviating temperatures. The curves XVII—XIX represent the metabolic rate of warm-blooded animals at body temperature and under basal conditions. Curve XVI represents BENEDICT's (5 and 6) average result of his studies on the hibernating marmot at a body temperature of 16° C, and curve XVII shows the respiratory rate of the same animal having a body temperature of 37° C. It should just be mentioned that the basal metabolism of the marmot seems to vary with the body temperature almost according to the standard curve of KROGH (59).

Only the curves XV, XVI, XVII and XIX of Fig. 46 represent

¹ For the N % used, cf. the text to the figure.

measurements of the true basal metabolic rate, while all other curves are average curves representing the metabolic rate during slight

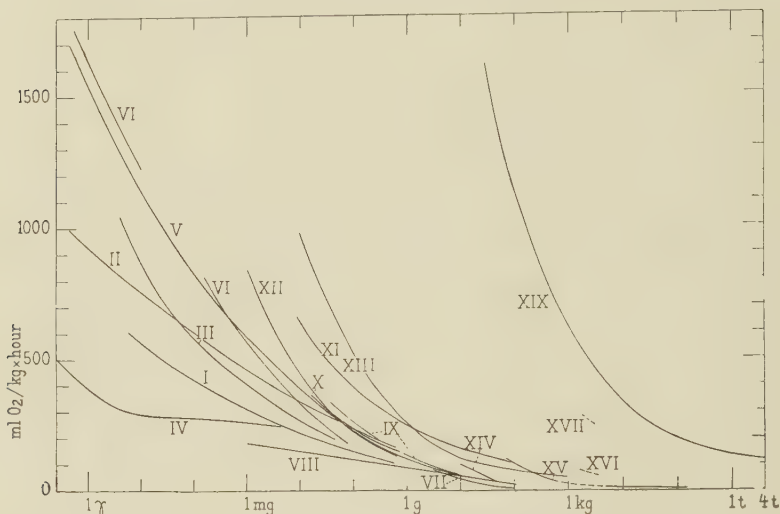


Fig. 46. Comparison of the metabolic rate of the animals on the basis of the body weight.

Ordinate: metabolism/weight. Abscissa: body-N.

The curves drawn are recalculated from those published in the present paper, viz.

- | | |
|---|--|
| I. <i>Vermes</i> , from Fig. 22, | 16° C. (2.0 % N). ¹ |
| II. <i>Littorina</i> , from Fig. 26, | 16° C. (2.2 % N). ¹ |
| III. <i>Nassa</i> , from Fig. 25, | 16° C. (2.2 % N). ¹ |
| IV. <i>Mytilus</i> , from Fig. 28, | 16° C. (1.65 % N). ¹ |
| V. <i>Crustacea</i> , from Fig. 30, | 16° C. (2.0 % N). ¹ |
| VI. <i>Balanus</i> , from Fig. 31, | 16° C. (2.25 % N). ¹ |
| VII. <i>Asterias</i> , from Fig. 33, | 20° (?) C. |
| VIII. <i>Vermes</i> , from Fig. 34, | 13-23° C. |
| IX. <i>Mytilus</i> , from Fig. 35, | 16° C. |
| X. <i>Gastropoda</i> , from Fig. 36, | 16° C. |
| XI. <i>Crustacea</i> , from Fig. 37, | 16° C. |
| XII. <i>Insecta</i> , from forest soil, | from Fig. 38, 13° C. |
| XIII. <i>Pisces</i> , from Fig. 40, | 16° C. (own exp.), 17° C. (Raffy), 20° C. (T. and D.). |
| XIV. Frogs, from Fig. 41, | 20° C. (T. and D.). |
| XV. Reptiles, from Fig. 42, | 20-25° C. (different authors). |
| XVI. Marmot, from Fig. 47, | 16° C. } (Benedict). |
| XVII. Marmot, from Fig. 47, | 37° C. } |
| XIX. Birds and mammals, from Fig. 43, | 37-40° C. (Benedict). |

¹ Own unpublished results.

(XIV) (and) or natural activity in so far as this state can altogether be defined (the rest of the curves). It may, furthermore, be mentioned that in Figs. 45 and 46 for the sake of clearness frequently only part of the curves are given. However, the conclusions drawn are unaffected.

Figs. 45 and 46 are much alike because the assumed N-percentages of the animals are rather uniform. The conclusions drawn on the basis of the results of Fig. 45 (1-4) can, thus, also be drawn on the basis of Fig. 46 and we shall, therefore, continue our comparison of the respiration intensities of the different groups of animals on the basis of the latter figure. We still compare animals of equal size, i. e. we compare "vertically" on the figure. It was mentioned under 3) that there is some indication of the fact that *Crustacea* respire more intensely than the equally large lower invertebrates and, 4) that fishes respire more intensely than do the equally large invertebrates including *Crustacea*. Fig. 46 indicates vaguely 5) that insects respire more intensely than do the *Crustacea*. It would seem as if 6) *Amphibia* and *Reptilia* respire rather less intensely than do the fishes. When comparing the hibernating marmot at 16° C (XVI) with the reptiles at 21° C, we must agree with BENEDICT (5) 7) that the warm-blooded animals, even at the same body temperature, respire considerably more intensely than their closest relatives, the reptiles. BENEDICT verified this conclusion by experiments in which he compared the metabolic rate of reptiles with that of mammals at a temperature corresponding to the body temperature of the latter animals. Thus, at all temperatures, mammals (and birds) seem to respire more intensely than reptiles. This is illustrated by Fig. 47 reproduced from BENEDICT's paper (5). The conclusion of the American author is perhaps somewhat debilitated by the theoretical considerations concerning the incubating, but otherwise resting, *python* whose body temperature and metabolic rate are definitely increased above the normal. It would, moreover, have been desirable in comparing warm- and cold-blooded animals that greater stress be laid upon a study of the metabolism of the lizzards, since snakes and especially tortoises are only remotely related with mammals (O. KUHN (67)). On the other hand, the marmot has the lowest basal metabolic rate of all equally large mammals studied and, for this reason, BENEDICT regards the hibernating mammals as a physiological unit which—with respect to the respiratory intensity—should be placed between the reptiles and the non-hibernating mammals. It seems thus

justified to conclude, in accordance with BENEDICT, that at the same body temperature and body weight 7) there is probably a considerable increase in the metabolic rate at the transition from reptiles to mammals and birds. Already in 1916, KROGH (60) held the same view. If the comparisons are made not at the same temperature, but at the "physiological" temperature (i. e. body temperature in the case

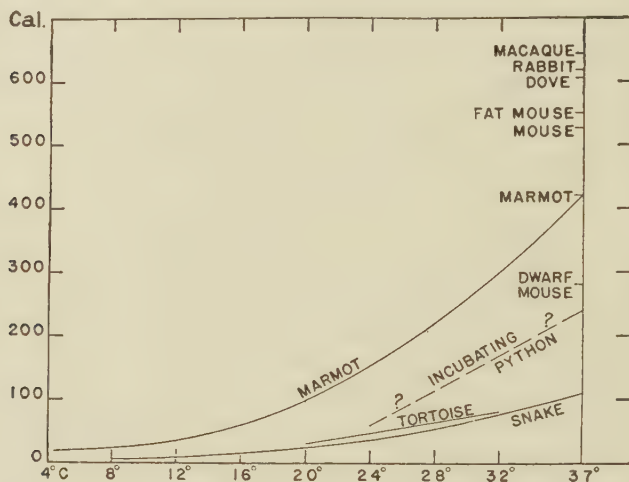


Fig. 47. Metabolism of different cold-blooded and warm-blooded animals, compared at a series of body temperatures. BENEDICT (5).

of warm-blooded animals and a reasonable air temperature in the case of cold-blooded animals), it is beyond all doubt that 8) a very marked "energy shift" takes place at the transition from reptiles to warm-blooded animals (at least a 20-fold increase in metabolic rate). It is reasonable to imagine that this "energy shift"—or at least "an energy shift"—has been of significance for the success of the warm-blooded animals in a world which once was dominated by the reptiles.

Figs. 45 and 46 illustrate 9) the consequent decrease in metabolic rate with increasing size of the animals throughout the animal kingdom. This decrease was found to prevail in all groups of animals studied. However, at the transition from one zoological group to another the metabolic rate of the animals may in some cases increase. This was indicated in the points 2–8 (excluding 6), as already mentioned previously.

Finally, it may be mentioned, 10) that only the smaller warm-blooded animals seem to reach an energy metabolism comparable with that of the marine micro-fauna. The average metabolic rate of the larger warm-blooded animals is comparable with that of the cold-blooded marine animals weighing 10–1000 mg (studied at 16° C). However, it should be kept in mind that most frequently the metabolic rate of warm-blooded animals is measured under basal conditions, while that of cold-blooded animals is generally determined during “normal activity”. It must be pointed out that the present comparison is based upon the study of a relatively minute number of species. With regard to any further generalization of the above statements some precaution should therefore be taken.

4.42. The Percentage Decrease in Metabolic Rate at a Ten Times Increase in Body Size.

If a cube with the edges 10 and the surface 600 is split in 1000 cubes with the edges 1, the aggregate surface of these small cubes will be 6000; hence, 10 times larger than the surface of the large cube. If RUBNER's law was generally valid, the metabolism/N or metabolism/weight should be reduced to $1/10$ whenever the body size of the animals increases by a factor 1000.

The Figs. 45–46 clearly indicate that the decrease in metabolic rate with increasing body size of the animals is in most cases considerably less than would correspond to the surface law. In the left part of both figures the metabolic rate generally decreases only to $1/2-1/4$ when the animals increase in size by a factor 1000.

However, we arrive at a better survey of these facts by analyzing the slope of every single curve throughout its total course and, subsequently, representing graphically the results obtained in a new figure. It was found most convenient to use the quantity “percentage decrease in metabolic rate at a 10 times magnification of the animals”. All results thus obtained are comprised in Fig. 48.

In this figure are given the results of the analyses of both the metabolism/N and the metabolism/weight curves. The single curves forming the basis of Figs. 45 and 46 have been used, because, in the two figures mentioned, it was found in some cases necessary slightly to displace curves which otherwise would have coincided.

In Fig. 48 the “percentage decrease in metabolic rate at a 10

times magnification of the animals" is plotted as ordinates. As abscissae are plotted, above, the N-content of the animals and, below, their weight. Both abscissae are in a logarithmic scale and the relative position of the abscissae was calculated on the basis of the assumption that all animals contain 2.2 % N. The error thereby introduced is without any significance, since Fig. 48 com-

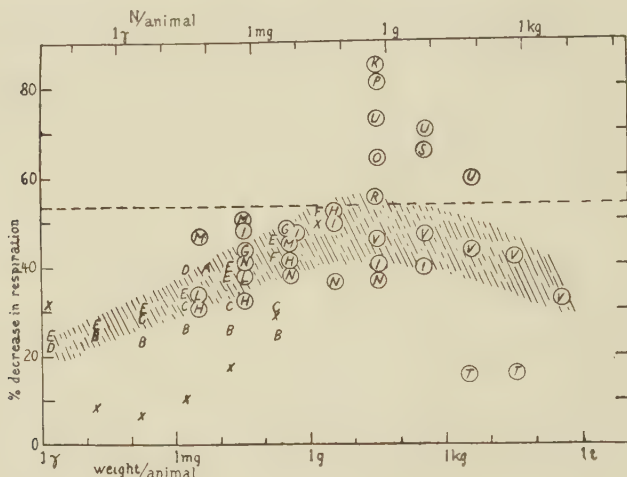


Fig. 48. The percentage decrease in metabolic rate for a 10-fold magnification of the animals (ordinate). Upper abscissa: body-N (results without circles). Lower abscissa: body-weight (encircled results). The results have been gained from the analyses of the slopes of the curves depicted in the following figures of the present paper:

The author's own results:

A	<i>Asterias</i> ,	Fig. 21
B	<i>Vermes</i> ,	" 22
C	<i>Nassa</i> ,	" 25
D	<i>Balanus</i> ,	" 31
X	<i>Mytilus</i> ,	" 28
E	<i>Crustacea</i> ,	" 29
F	<i>Pisces</i> ,	" 32
G	<i>Gastropoda</i> ,	" 36
H	<i>Mytilus</i> ,	" 35
I	<i>Crustacea</i> ,	" 37

From the literature:

K	<i>Asterias</i>	Fig. 33
L	<i>Tenebrio</i>	" 39
M	Insects from forest soil	" 38
N	<i>Pisces</i>	" 40
O	Frogs (BLANK)	" 41
P	Frogs (T. and D.)	" 41
R	<i>Lacerta</i> (KRAMER)	" 42
S	Reptiles (T. and D.)	" 42
T	Large reptiles (BENEDICT)	" 42
U	Birds (GROEBBELS) (34), figure drawn by the author, unpublished	
V	Birds and mammals	" 43

prises animals the body size of which varies by a factor 10^{12} (from the *Mytilus* gastrula to the elephant). The points obtained from the metabolism/weight curves are surrounded by circles, those from the metabolism/N curves are without circles.

The single results show a marked scattering but, nevertheless, it is clear that the "percentage decrease in metabolic rate" with increasing body size of the animals is definitely less pronounced for the small than for the larger animals, the average trend being that of a gradual increase of this value with increasing size of the animals. The subsequent diminution in "percentage decrease in metabolic rate" in the left part of the figure describing BENEDICT's experiments on very large animals is mainly based on a few experiments on elephants and should perhaps not be taken very serious. BRODY pointed out that the metabolism of adult birds and mammals—as compared interspecifically—is most satisfactorily described by the formula $Q = 70.5 \cdot M^{0.73}$. In the nomenclature applied here, this would mean that the metabolic rate of the warm-blooded animals over the whole size range of these animals decreases by 45 % each time the animals increase in weight by a factor 10. BRODY mainly utilized BENEDICT's figures and already in 1935 the "Committee of Animal Nutrition of the National Research Council" adopted his view (quoted from (5) and (14)).

Considering the whole material published in Fig. 48 the present author regards it as justified to lay most stress upon the shaded area, especially when taking into account his own and BENEDICT's results.

It may thus be emphasized that the author's own experiments together with information gained from the literature are very much in favour of the view that the metabolic intensity of the animals decreases most markedly with increasing body size in the range of medium size animals, however, gradually less for smaller and, maybe, also for larger animals.

By simple calculation, we may arrive at the following: if, generally, the metabolic rate of the animals would follow the "surface law" of RUBNER, the metabolic rate of animals should decrease by c. 52 % whenever the body size increases by a factor 10. Therefore, the surface law is expressed by the horizontal, dotted line of Fig. 48.

If we keep in mind that the shadowed area of Fig. 48 represents

an average only, it becomes clear that, especially in the case of medium size animals, we shall frequently find a rather close interdependency between metabolism and surface. Notably, the animals of medium size have primarily been the object of size-metabolism investigations. Assuming the surface law to be generally valid, attempts have been made (PÜTTER (93)) to calculate the metabolic rate also of the smallest and largest animals. In the case of bacteria and other micro-organisms, PÜTTER calculated (and found as the result of imperfect measurements) in this way astronomically high figures. For animals PÜTTER's figures do not agree with reality, i. a. because the metabolic rate of the animals does not follow the surface law, as it was supposed by him.

The account given in the preceding theoretical part of the present work (Section 4) must unfortunately be very subjective. However, the author considered it impossible to subject the figures and the curves to any kind of probability calculation. For such calculations the present material has been much too heterogeneous. However, according to the author's view the evaluations given will gain in probability as soon as the data forming their basis can be arranged within some kind of system or order. The Figures 45, 46, and 48 seem to reveal such a system, the possible basis of which will be discussed in the following.

5. DISCUSSION

5.1. Metabolism and Food Requirements.

Provided that in the animals studied in this work combustion is complete, as it is known to be the case in mammals (an assumption which is by no means proved), this would mean that the oxygen consumption must be an approximate measure of the calory production of the animals and, hence, also of the quantity of food absorbed through the alimentary canal. On the average, a 10 γ -animal would then combust 5 times as much food per weight- and time unit as does a 1 g-animal (cf. Fig. 46).

However, the calory production is no measure of the quantity of food ingested, because some food is used for growth and, moreover, some food is probably not at all absorbed in the intestine but is again lost with the feces. For those animals of the present paper, which have been studied over their total size range, *viz.* from larvae to adults, we may generally assume the growth rate (defined as the

percentage increase in weight per time unit) to be the higher the smaller the animal; and, moreover, there is good reason to assume that the utilization of the food in the intestine is the poorer, the shorter the intestine of the animal in question, i. e. the smaller the animal.

Both factors mentioned above, therefore, tend to increase the food requirement of the micro-animals, as compared with that of the macro-animals, beyond the quantity calculated from the metabolic rates. Possibly, this increase can become very considerable, as is indicated by comparing the author's metabolism experiments with the filtration studies of BARKER JØRGENSEN (47). On beforehand, one would expect the filtration rate to be a measure of the quantity of food held back by bivalves; BARKER JØRGENSEN's survey of the literature available shows that the filtration rate in the oyster larva (i. e. filtration calculated per unit of time and body weight) is about 600-700 times larger than the rate of filtration in a very large *Mytilus*. The author's metabolism experiments held together with BARKER JØRGENSEN's filtration studies seem to indicate that the ratio $\frac{\text{food filtered off}}{\text{food combusted}}$ is about 100—200 times higher in the larvae than in the old bivalve animals. Thus, it becomes a question of greatest interest and maybe of utmost importance, where the undigested food in the larva takes its way (detritus which sink to the bottom?). The rate of filtration in larger bivalve animals (FOX's experiments) has presumably been measured too low, however, and there is good reason to believe that the weight of the larger animals was estimated too high, as compared with that of the larva (own unpublished observations). Both facts tend to make the enormous difference between the rate of filtration in small and in large animals smaller than stated above. However, the suggestive differences between BARKER JØRGENSEN's and the present author's results must be considered to persist.

5.2. *Metabolic Rate and Body Size.*

5.21. Introduction.

On the basis of the experimental material available until the present it is attempted to gain a picture of how the metabolic rate of animals depends on their body size. In order to arrive at general rules, the whole animal kingdom ought to be surveyed, but only a

minute number of animals have been studied. Therefore, any generalization must necessarily become very uncertain. Accordingly, at the best, the value of the present study will be its showing the way for further investigations.

As regards the smallest animals, the experiments available have been performed exclusively on marine cold-blooded animals (the author's experiments) occurring mainly in the surface layer of the Sound where the salt concentration varies considerably (usually it is 11-14 ‰, according to THORSON (109)). Concerning animals of medium size, both cold-blooded marine, limnic, and terrestrial forms are dealt with, while the large animals discussed are preponderantly warm-blooded, terrestrial animals.

Actually, on the whole the small animals studied are more primitive than the larger ones which have been the objects of investigation. Primitive is here used in the sense that the animals in question belong to lower parts of the zoological system. Hence, the coarseness of the above statement is obvious. Since life spread from the sea mainly via fresh waters to the land and to the air, it appears that the sequence from small to larger animals at the same time roughly indicates the direction of the evolution of the animals studied both as regards the zoological system and the milieu. Some authors state that the metabolism of the animals is largely dependent upon their living habit and upon the biotop upon which they live. The experiments of SPÄRCK (103) and also those of FOX and SIMMONDS (29) showed the mentioned dependence. The British authors found a 1.5-3 times higher metabolism of limnic crustaceans as compared with related marine crustaceans. Their experiments were, however, performed on anaesthetized animals and no proof has been given that all the animals studied are equally sensitive to the anaesthetic employed.

However, the experiments mentioned show the necessity of extending our future studies to a much larger number of specimens. The oceanic, the limnic, and the terrestrial micro- and macro-fauna would be suitable objects of experiments which would be directly comparable with the author's own experiments on marine animals.

5.22. The Author's Own Investigations.

The author's experiments showed the micro-fauna of the sea (animals weighing around 10 γ) to respire on an average 5 times

more intensely than the 10^5 times larger marine animals weighing around 1 g. A comparison of animals varying still more in size reveals still greater differences of the respiration intensity.

In the micro-experiments the Cartesian diver was used, and in the macro-experiments the oxygen consumption was determined according to the Winkler principle. All experiments were performed during the summer time from early May to the second half of August. The micro-experiments were made in 1944 and the macro-experiments in 1946. In all cases, the temperature of the experiments was 16°C , but on the average the macro-experiments were carried out 4-6 weeks earlier in the year than were the micro-experiments. Therefore, the temperature of the sea was somewhat lower ($3-4^{\circ}$ on the average) in the case of the macro-experiments. Some orientating macro-experiments, however, were performed in 1944 simultaneously with the micro-experiments.

In view of the fact that the micro- and macro-experiments have been performed with a different technique and at a (slightly) different time of the year, there may be some reason to discuss whether they are quite comparable.

The highest possible influence of the mentioned time difference on the majority of the micro- and macro-experiments may best be estimated on the basis of the experiments with crustaceans (cf. p. 99). The reason is that only in the case of these experiments a definite difference between the orientating macro-experiments from 1944 and the final ones from 1946 could be observed. On the average, the metabolic rate of the 1946-animals seems to be 40% higher than that of the 1944-animals. It therefore seems reasonable to assume that generally the difference in metabolic rate of 40% is the maximum influence of the time and the temperature difference mentioned upon the metabolism of the animals under investigation, when compared at one temperature. These 40% would appear to be a rather high estimate in view of the small differences in calendar and in temperature; this also becomes apparent from the works directly dealing with the significance of the temperature at which the animals have previously lived for their metabolic rate. On a teleostean fish, *Gallichthys mirabilis*, WELLS (113) found the metabolic rate (measured at 22°C) to be 50-100% higher in individuals acclimatized to c. 11°C than in individuals acclimatized to c. 33°C . On the crab *Emerita talpoida* EDWARDS and IRVING (26) found the metabolic rate of the winter animals (from -1.5°) to be only about 30% higher than the

metabolic rate of the summer animals (from 20-21° C), both groups of experiments being performed at the same temperature (16° C). Only when compared at very low temperatures (3° C) the difference is considerable, the metabolic rate of the winter animals being several times higher than that of the summer animals. On the sand flea *Talorchestia megalophthalma* the same authors found no difference between the metabolic rate of the winter- and summer animals when compared at different temperatures and, finally, on the teleostean fish *Tautoglabrus adspersus* HAUGAARD and IRVING (38) found only small and unreliable differences between the metabolic rate of summer- and winter animals when compared at the same temperature. The two last mentioned animals were from the same locality as was *Emerita*.

The question whether the diver method leads to results which are systematically different from those obtained with the Winkler method is difficult to answer. In both cases, about the same time elapsed between the catching of the animals and the commencement of the experiments. After the initial handling and prior to the experiments, the animals spent about the same time, *viz.* 1 hour, in the diver and in the flask, respectively. In the flasks, however, the water had to be renewed immediately before the start of the experiments; this was done with the greatest caution but, of course, the procedure must have disturbed the animals and, hence, the metabolic rate of the macro-animals during the measuring period may have been a little increased (in the flasks) as compared with that of the micro-animals in the divers. Thus, for example, HAUGAARD and IRVING (38) performed manometric measurements of the oxygen consumption of the fish *Tautoglabrus* and they mention that during the first 10-30 minutes after the animals had been placed into the apparatus the metabolism is slightly increased, but later it becomes quite constant. However, the figures published show that, when the measurements are performed during 1-2 hours' periods, we shall find the metabolism in the first period after the start of the experiment to be only some 10% higher than in later periods. Obviously, these results cannot directly be applied to the present author's experiments, but it seems permissible to assume that not either in the present experiments have the disturbing effects been essentially higher. At any rate, the author did not observe any reliable differences between flask experiments extended over a shorter or a longer period of time.

On one single object, *viz.* plankton copepods, only, have experiments been carried out both in flasks (6 experiments) and in divers (5 experiments). The results of diver experiments on the copepods *Centropages* and "unknown" are on an average 25% lower than the results obtained in flask experiments with mixtures of copepods (cf. Figs. 29 and 30). The difference found may possibly be ascribed to one or to some of the following factors. 1) The probably slightly smaller size of the animals in the flask experiments, 2) the diver experiments were carried out about 2-3 weeks later in the year than were the flask experiments, 3) species-differences of the animals, 4) owing to the above mentioned disturbance effect, the flask experiments may have led to somewhat higher results. In all, the control experiments on copepods do not seem to support the presumption that flask- and diver experiments lead to considerably deviating results.

It has been mentioned that the ratio between the metabolic rate of animals weighing about 10 γ and that of animals weighing about 1 g averages 5 : 1. Perhaps this ratio will not turn out to be quite true but, at any rate, it will scarcely exceed the value 8 : 1, and the correct ratio lies presumably between 5 : 1 and 8 : 1. Even if one assumes the ratio 8 : 1 to be correct, this will only to a minor degree affect the curve analyses given in Section 4.42. A decrease in metabolic rate from 5 to 1 at an increase in body size from 10 γ to 1 g (10^5 times) means that whenever the body weight of the animals under consideration increases by a factor 10, the metabolic rate decreases by 27%. Thus, the ratio 8 : 1 corresponds to a decrease in metabolic rate of 35% whenever the animals grow 10 times larger. This percentage is still considerably lower than the average of 50% found for still larger animals (cf. Fig. 48). If the respiration intensity would decrease by 50% as the body weight increases 10 times, the 10 γ animals would respire 32 times more intensely than the 1 g animals. Finally, the surface law ($\sim 52\%$ as defined above) claims that the metabolic rate of the small animals considered should be 40 times higher than that of the larger animals studied.

Even if we take into consideration possible errors in the comparison of the respiration of small and large marine animals, the errors can hardly be so great that they can upset the general conclusion of the present treatise, *viz.* that animals weighing less than 1 g most frequently do not obey the surface law. The metabolic

rate of the smallest animals investigated was systematically higher than that of larger animals, however, not nearly so high that the surface law could be applied.

5.23. Metabolic Rate Variations throughout the Life of the Individual Animal. The Possible Bearing of this Phenomenon on the General Understanding of the Metabolic Rate-Body Size Relations.

Fig. 48 shows a simple analysis of the slopes of the most important curves comprised in the present work. The analysis makes it clear that for practically all animals studied the metabolism per weight or per N decreases with increasing body size. On the average, the percentage decrease in metabolic rate at an increase in size of the animals by a factor 10 is lowest for the very smallest animals, highest in the range of medium size animals, and maybe again somewhat lower in the case of very large animals.

Fig. 48, thus, faintly indicates an interrelationship connecting animals from the whole animal kingdom and manifesting itself through the rate of decrease in metabolic rate with increasing body size of the animals. The existency, however, of such an intricate relationship between body size and metabolic rate throughout the whole animal kingdom is very difficult to understand and, hence, also very difficult to accept. The surface law expressing a dependency between metabolism and $W^{2/3}$ has, thus, been replaced by a less simple, perhaps more correct law which does not, however, seem to be easier to understand than was the surface law.

When we consider Fig. 48 and the curves (Figs. 45 and 46) from which it has been derived, it becomes evident that the material to be compared is very heterogeneous. Part of the curves have been obtained by interspecific comparison of the animals, i. e. by comparing the metabolic rates of related, small and large species. Other curves were gained by intraspecific comparisons, i. e. by comparing the metabolic rates of individuals of different size, however of the same species. In this latter case, the results were obtained on animals of widely different ages and growth rates, the small animals always being younger and usually growing much more rapidly than the larger animals. If, nevertheless, intraspecific and interspecific comparisons have not been kept apart in the present work, this is

due to the fact that in his own experiments the author has been unable to detect from the beginning any definite difference in metabolic rate between small adult and small larval worms and crustaceans.

In the following, however, we shall distinguish sharply between intra- and interspecific comparisons and, at this place, we will state that each interspecific curve must be regarded as having been obtained by a comparison of a large number of generally very small parts of intraspecific curves. The complete course of the intraspecific curve is as a rule unknown to us. In a few cases, however, it will be possible to gain an impression of the metabolic intensity throughout the whole ontogenesis of a species. These few cases will be discussed below.

The variation of the metabolic rate with the body size has in no case been studied very exactly over the whole size range of a species. The *Mytilus* curve (Fig. 28) represents the most complete approach. This curve (apart from the part to the utter left) shows a steeper and steeper decrease in metabolic rate with increasing body size of the animal. In the case of the plankton larvae and of the smaller sedimented animals, the metabolic intensity is almost independent of the body size, but on the larger sedimented animals we see a continuously steeper decrease in metabolic rate. In the case of *Mytilus*, the convexity of the curve turns upward. Even in the case of the concavity turning upwards, however, the decrease in metabolic rate, expressed in per cent, may very well increase with increasing body size of the animals. This holds presumably for *Nassa* (Fig. 25) and for *Littorina* (Fig. 26). The *Asterias* curve (Fig. 21) seems to show that, in the case of the very large starfishes, the metabolic rate decreases only slightly with increasing body size. On the other hand, HELGA MEYER's experiments (Fig. 33) which are included (dotted line) in Fig. 21 just indicate a very steep decrease with increasing body size of the adults. The author has greater confidence in HELGA MEYER's thorough experiments than in his own few ones, especially since in two cases on very small *Asterias* (2–3 mg N) the author found a relatively high value of respiration/N. However, these experiments were unsatisfactory and they are, therefore, not published here, although the approximate correctness of the results can hardly be doubted. It seems, thus, possible that the metabolic rate of *Asterias* decreases only slightly with increasing body size in the case of small animals, but more markedly in the range of larger animals. Considering the vertebrates, we find in the *amphibia* (Fig. 41) a marked decrease in metabolic rate with increasing body size (fat curves). However, when including GROEBBELS' results on small, just metamorphosed frogs, it appears extremely probable that the decrease in metabolic rate with increasing body size is considerably less for very small frogs (dotted curve). It may also be mentioned that WILLS found the metabolic rate of tadpoles not to be essentially different from that of the adults (quoted from BRACHET (13)).

We know that the metabolic rate of the *mammalia* decreases markedly with increasing body size of the adults, and it is known from numerous experiments (cf. BENEDICT (5)) that this holds both for mammals as a whole and for the single species. No data, however, are available which permit the metabolic rate of a single species to be followed throughout its whole development from egg to adult.

It should be emphasized, however, that assuming the metabolic rate during embryonic life to decrease with increasing body weight in the very same manner as is known from adult mammals (O_2 consumption $= k \cdot W^{0.73}$) we shall arrive at improbably high metabolic rates for the very young foetuses. For example, a foetus weighing 10 g would respire about 100 times more intensely than a fully grown mammal weighing 1 kg. The very few informations available in the literature, however, are not at all in favour of this view:

BOELL and NICHOLAS (9) state that a rat embryo in the 2–8 cell stage consumes $0.00073 \mu l O_2$ /egg and hour. On the 8th day of the development, the same value is $0.01 \mu l$ /foetus and hour and, on the 10th day, it is $0.2 \mu l$ /foetus and hour. The dry matter of an egg is stated to weigh 0.02 – 0.033 g. From PINCUS' measurements (90) of the egg diameter (0.07 – 0.075 mm) the egg volume may be calculated to 0.18 – $0.22 \cdot 10^{-3} \mu l$ (mean $0.2 \cdot 10^{-3} \mu l$). Thus, the eggs of BOELL and NICHOLAS can be assumed to contain 10–17 per cent dry matter. In the cleavage stages of the rat egg the metabolism/wet weight can thus be calculated to be 3000–4000 ml O_2 /kg and hour. The American authors maintain that "when embryonic growth occurs the oxygen consumed accurately reflects the increase in embryonic mass". Presumably, this means that the metabolic intensity remains constant during the first 10 days of development. However, it is not clear whether this holds for the metabolic rate as calculated per dry weight or per wet weight. BRACHET ((13), p. 276) holds the view that metabolism per "weight of the embryo" (wet weight?) during the period in question decreases to about half the metabolic rate just after fertilization (according to PINCUS (91) a ten days old foetus is 1.0–1.5 mm long). BRACHET seems to have had access to informations which are not available for the present author. In a later communication (10) BOELL reports new experiments showing low respiratory rates for rat embryos containing about 3 g N. ($8.5 \times 10^{-2} \mu l O_2$ per g N and hour (comp. Fig. 46)).

On a rat foetus weighing 77 mg KLEIBER et al. (cit. from BARRON (2)) found a metabolic rate of 710 ml/kg and hour. The respiration of the adult rat (400 g) amounts to c. 600 ml/kg and hour (cf. Fig. 43). However, it should be kept in mind that adults and embryos are not directly comparable, because after birth the metabolic rate always increases somewhat.

The figures available from experiments on rat foetuses, thus, indicate that the metabolic rate decreases from 3000–4000 to 710 when the mass of the foetus is increased by a factor 400000.

This corresponds to a decrease in metabolic rate of only 22 % ($\propto W^{0.9}$) at an increase in mass by a factor 10. For adult animals, the same value is c. 45 % ($\propto W^{0.73}$). The author is fully aware of the fact that the water content of the embryos may fluctuate and that this may affect the accuracy of the calculations; however, this cannot be very important, since the water content

of the eggs (10–17 %, 15 % according to BOELL (10)) can hardly be very different from that of KLEIBER's fetuses.

Finally, it may be mentioned that BARCROFT's experiments on sheep embryos showed an almost constant metabolism/weight when the weight of the embryo varied between 0.8 and 3.6 kg (quoted from WINDLE (116)).

It seems reasonable to conclude that also in the case of mammals further experiments will probably show the percentage decrease in metabolic rate to increase as the individual grows. This presumably also holds for the metabolism calculated per N of the animals, such as it will be demonstrated below in the case of birds.

Experiments on bird embryos (PHILIPS (91)) showed that during the first 17 hours of development (growth and measurement at 38° C) the metabolism/N of the isolated blastoderm increases from 3.3 to $7.5 \cdot 10^{-2}$ μ l O₂/g N, hour. In the following 20 hours, and maybe for another 2 days, the metabolic rate remains constant. Subsequently, we may no doubt expect the metabolic rate to decrease. An isolated 17 hours old embryo contains circa 4–7 g N. It appears that the chicken embryo at 38° respire almost twice as intensely as the equally large marine animals at 16° C. If measured at 16° C, the metabolism/N of the chicken embryo would undoubtedly have been found even lower than that of the bulk of the marine micro-fauna.

The bird embryo does not respire definitely more intensely than does the adult bird. Hence, during embryonic life, the decrease in respiratory rate with increasing body size must be very slight or even approach zero. This agrees well with the findings of PHILIPS and it has also been emphasized by BRODY (14, p. 430).

The above survey is very much in favour of the view that for the single species of cold-blooded and of warm-blooded animals the metabolic rate quite generally shows a steeper and steeper percentage, and in some cases even absolute decrease as the individual during its ontogenesis gradually grows larger.

It should, however, be mentioned that exceptions from this rule have been reported. Thus, WHITNEY (115), studying four species of planarian worms, found the metabolic rate of three species to decrease with increasing body size, while the metabolic rate of the fourth one increased with increasing body weight. However, the size variation of the last mentioned species was only small, namely from 2 to 10 mg.

The question now arises, whether very small species and considerably larger ones actually deviate from each other with respect to the way in which the metabolic rate decreases with increasing body size. As a matter of fact, the present author recognizes differences which seem to indi-

cate that the metabolic rate first decreases markedly when the species approaches the upper limit of its size range. It seems to be of minor importance whether this limit is high or low. In the crab *Emerita talpoida* (EDWARDS and IRVING (26)) the metabolic rate decreases by about 90 % when calculated for a ten times magnification of the animals near the upper size limit of the species. However, the animals have been studied over a narrow size range from 0.3 to 0.8 g only, but the experiments are very conclusive. The metabolic rate of *Asterias* (MEYER, Fig. 33 of the present work), of *Ascaris* (KRÜGER (65)), and of frogs (TERROINE and DELPECH, BLANK, Fig. 41 of the present work) show a marked decrease (almost 80 %) at an increase in body weight of the respective animals by a factor 10. The animals studied all approach their upper limit of body size, i. e. here about 10-100 g. BENEDICT's experiments (3) on large reptiles weighing 1-100 kg showed most frequently that within one and the same species the metabolic rate decreases definitely with increasing body size. BENEDICT (p. 470) emphasizes that the surface law is approximately valid, with the only exception of the large tortoises, i. e. he finds the metabolic rate to decrease by c. 50 % when the body weight increases 10 times. When comparing the large reptiles from species to species, however, we find only about 15 % decrease in metabolic rate for every 10-fold increase in body size (cf. Fig. 42, III).

Unfortunately, considerable uncertainty is prevailing concerning the reptiles. Experiments performed on other groups of animals (for example, insects, fishes) are so contradictory that a closer analysis would appear to be of minor value. On the basis of the material already given, however, the author considers it worth while to emphasize that the metabolic rate often decreases most steeply with increasing body size as the species in question approaches its largest possible size, in other words, as growth (maybe due to genetical or endocrine factors) gradually ceases. This phenomenon appears to be independent of whether the respective species is large or small.

After the conclusion of the present manuscript the author received BRODY's book (from 1945) dealing mainly with domestic animals, the metabolic rate of which was studied throughout the whole period of growth (excluding embryonic life). For each species he reports a steeper and steeper decrease in metabolic rate with increasing body size. This is in complete agreement with the findings of the present work, valid for those species where also

the embryonic period was included in the investigation (*Mytilus*, frogs, mammals, and birds). Thus, BRODY's comprehensive experimental material (14) strongly supports the above views.

It should now be stressed that in very few cases, only, the present author has made an attempt to determine the relationship between metabolic rate and body size within the narrow range near the maximum size of the respective species. In the present experiments, a considerable variation in size of the animals in question has been obtained by comparing adults with larvae of the same species or with smaller or larger adults of related species. Almost all other workers obtained a reasonable size variation by including in their intraspecific comparisons individuals of a maximum size. Thereby, a certain discrepancy is prevailing between the author's material and most other intraspecific comparisons described in the literature. The present author, therefore, wants to emphasize the possibility that steep decreases in metabolic rate with increasing body weight might also be found by intraspecific comparison of very small species, for example, small copepods. However, such experiments are still lacking; when at hand, they possibly will to a considerable extent blur out the general picture of Fig. 48. For this reason, Fig. 48, as it is given here, may in the future appear to be the result of the analysis of an inhomogeneous material.

Finally, it should be mentioned that—according to Fig. 46—the metabolic rate of the marine micro-animals varies rather considerably. But considering, for example, organisms weighing circa 10 γ the metabolic rate at 16° C in most cases appears to fall within the limits of 500-1500 ml O₂/kg, hour. Now, according to BOELL and NICHOLAS's experiments, very young rat fetuses of almost the same weight consume circa 3000-4000 ml O₂/kg, hour at 37° C, i. e. 3-4 times as much. If the metabolic rate of the rat fetuses had been measured at 16° C, their oxygen consumption would probably not have been found to deviate markedly from the metabolic rate of equally large marine animals. The same argument holds when we compare KLEIBER's experiment at 37° C on a rat fetus weighing 77 mg with experiments on equally large marine animals at 16° C. Had the comparison been made at the same temperature, we would

hardly have found large differences in oxygen consumption. It has already been mentioned on p. 143 that in early stages of development the metabolism/N of bird embryos would most probably have been found even lower than that of most marine animals of the same size, provided the experiments had been performed at the same temperature.

We may, therefore, state that the metabolic intensity of all animals studied, when measured at the same temperature, seems to scatter around a characteristic value, and it appears to be of minor importance whether the animals considered are small adult animals or they are larvae or embryos from all regions of the animal kingdom, the only condition being that the larvae or embryos do not contain appreciable amounts of yolk, the presence of which obviously might markedly lower the values of respiration/N or respiration/weight.

It has been mentioned previously that every interspecific curve relating body size with metabolic rate may be regarded as being composed of a large number of intraspecific curves, the complete course of which usually is unknown to us. The most marked decrease in metabolic intensity with increasing body size of the animals has been found in intraspecific comparisons and, generally, in a region around the greatest possible size of the species investigated (*Asterias*, *Ascaris*, *Emerita*, frogs), and never while the animals still were in earlier periods of growth. Within one and the same group of animals, the interspecific comparison, therefore, may often show a slighter decrease in metabolic rate with increasing body size of the animals than found in intraspecific comparison of the different species involved (large reptiles). These observations induced the author to consider the possibility that the metabolic rate of the animals throughout the animal kingdom might be related in a manner which may roughly be illustrated by the scheme of Fig. 49.

For the sake of clearness, only three groups of animals are included in this scheme, *viz.* two cold-blooded (I and II) and one warm-blooded group (III). The weight of the animals is given on the abscissa (logarithmic scale) and the metabolic rate (calculated per unit of weight) is plotted equidistantly on the ordinate. Throughout the period of growth, the metabolic rate of the warm-blooded animals is higher than that of the cold-blooded animals. When the

comparison is made at 16°C , however, the metabolic rate of the embryonic warm-blooded animals hardly differs from that of the equally large cold-blooded animals. This is indicated by the dotted line III'. In order to facilitate the survey of Fig. 49, curve II has not been drawn back to the common origin of the curves.

Curve I is intended to illustrate the possible relations prevailing in a group of small cold-blooded animals, and curve II shows the

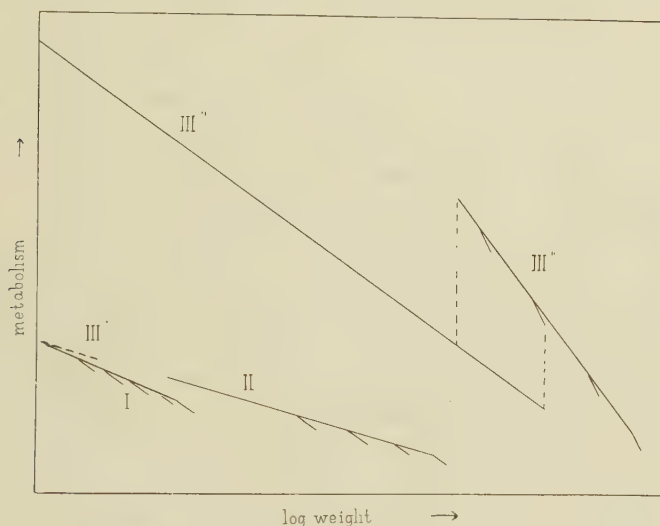


Fig. 49. Scheme illustrating the probable evolution of the metabolic rate in animals.

same for a group of rather large cold-blooded animals. Generally, the metabolic rate decreases with increasing body size of the animals; this is shown by the straight lines drawn. These lines also indicate that the percentage decrease in metabolic rate becomes steeper and steeper with increasing body size. Whenever a species approaches its final size, the decrease becomes still steeper, and this is in each case indicated by a short slanting line. Also in the case of warm-blooded animals having the body temperature $37\text{--}38^{\circ}\text{C}$, the metabolic rate decreases first slowly with increasing size of the foetus (III'') and then more rapidly with increasing size of the young ones and of the adults (III'''). An increase in respiration at the transition from embryonic to adult life has been indicated in the figure (III'' and III''') corresponding to the fact that the metabolic rate increases after birth (mammals) or after hatching (birds). Maybe the

curve illustrating the metabolic intensity during embryonic life of warm-blooded animals (III'') should have been drawn flatter, thereby indicating that the metabolic intensity in some cases remains rather constant throughout periods of considerable growth (PHILIPS (91), BRODY (14)).

If the views illustrated by Fig. 49 in the main points are correct, all metabolic rate-body size relations can be comprehended under one main aspect: the change in metabolic intensity throughout the life of the individual.

The steeper and steeper percentage decrease in metabolic rate with increasing body size of the animals throughout the whole animal kingdom (Fig. 48) may now quite simply be explained by assuming the metabolic rate generally to decrease more and more as the growth period of the single species proceeds.

The left part of Fig. 48 represents intraspecific comparisons of organisms in full growth, or interspecific comparisons of organisms which partly still grow. As it appears from Fig. 49, both ways of comparison will show low percentage decreases in the metabolic rates of animals. Around the middle of Fig. 48 the intraspecific comparisons dominate; according to the present author's view, they were most frequently performed on animals whose growth has slowed down. According to the scheme of Fig. 49 this should involve a high percentage decrease in metabolic rate. In fact, this seems to be so. In the right part of the figure we find mainly interspecific comparisons performed on animals whose growth has almost come to a stand-still (adult crustaceans, birds, and mammals). On the average, we here find a moderate decrease in metabolic rate, which is also in agreement with the scheme of Fig. 49.

The important figure on p. 372 in the book of BRODY (14) shows that in the case of mammals the metabolic rate generally decreases more and more steeply with proceeding growth, i. e. from birth to fully grown animals. When comparing equally large growing and fully grown animals, BRODY's figure shows that the metabolic rate of the growing animals is slightly (on the average c. 25 %) higher than that of equally large adults. Also this is, thus, in agreement with the scheme proposed in Fig. 49.

Hence, the tendency revealed in Fig. 48 must be considered to result from two facts, viz. 1) the experimental material underlying the figure is heterogeneous and, 2) generally, the metabolic rate decreases more

and more rapidly during the whole ontogenesis of the single species.

An especially steep decrease in metabolic rate in the proximity of the largest size of the species in question makes us assume the curves to be different for large and for small species. This view involves the important—and to the author's opinion acceptable—consequence that, with respect to energy metabolism, the species specificity of an organism becomes manifest only at the end of its ontogenesis, when the process of growth gradually stops.

5.24. Why Does the Metabolism per Unit of Weight or N Decrease with Increasing Body Size of the Animals?

For the past hundred years or even longer, this problem attracted the interest of the biologist. However, a question like this can hardly ever be answered definitely and, so far, the discussion has not contributed very much to the elucidation of the problem. Primarily, the discussion centered around the relation between metabolism and body size of larger animals. RUBNER found proportionality between body surface and metabolism in dogs ("surface law") and, in the first place (98, 99), he considered the results to be the necessary consequence of the heat regulation in warm-blooded animals. Later, however, he modified (100) his view, regarding the "surface law" as an adaptation to the conditions of life usual for homeotherm organisms. Most authors, *viz.* KESTNER (48), KRAMER (55), TERROINE and DELPECH (106), and BUDDENBROCK (19), however, cannot accept RUBNER's interpretation, because also in many cold-blooded animals the metabolism varies almost proportionally with the surface. Finally, BENEDICT and coworkers (5) in very accurate experiments showed that, as a matter of fact, only rather few warm-blooded animals follow the "surface law"; for most species, the deviations from this law are very marked. Comparing the species mutually, these authors definitely established that the warm-blooded animals as a whole obey a law which deviates considerably from the "surface law".

For this reason, the present author, in agreement with RUBNER's numerous critics, considers it reasonable to explain the decrease in metabolic rate with increasing body size of the animals by factors which have nothing to do with heat regulation. Evolutionary, heat regulation is of a very recent date and, presumably, some kind of interdependency between metabolism and body size has by primitive

warm-blooded animals been taken over from poikilotherm ancestors. Such original relationships may well have become modified in the course of the further evolution of the homoiotherms.

In the literature, attention has been focused upon several factors which make the metabolic rate of large animals higher than that of small animals. Of special importance are the experiments performed in order to determine in which way the weight of internal organs (heart, kidney, liver), showing a very high metabolic rate, varies with the size of the animals. TERROINE and DELPECH (106), GROEBBELS (32, 33), KESTNER (48), and LIANG, CHUNG MOU (70) showed that these organs are relatively larger the smaller the animal. Evidently, this must contribute to the fact that small animals show a higher metabolic rate than larger ones but, as has been emphasized by BUDDENBROCK (19), the higher the metabolic rate of an organism, the larger internal organs will be required. The phenomenon can, therefore, well be of a secondary nature relative to the primary one: the high metabolic rate of small animals, which we want to explain. (However, BUDDENBROCK does not mention that the work performed by internal organs may be higher in small than in large animals because, presumably, the metabolic rate of the organs proper is higher in the former than in the latter; cf. the cited paper (52)).

KESTNER (48) introduced the denotation paraplasma for the "skeleton-proteins" of the cells, while the term protoplasma was reserved for the respiring proteins. KESTNER's pupils BLANK (6) and LIANG, CHUNG MOU (70) searched, however with negative result, for differences in the ratio $\frac{\text{paraplasma}}{\text{protoplasma}}$ by studying the ratio $\frac{\text{trypsin digestible N}}{\text{trypsin undigestible N}}$ of smaller and larger animals (frogs, reptiles, mice). These experiments, therefore, gave no support to KESTNER's assumption that the non-respiring skeleton-protein should increase more than the respiring protein proper with increasing size of the animals. However, this possibility cannot at this time be excluded. VOIT (quoted from KROGH (60)), on the basis of similar considerations, laid special stress upon the determination of the "Organeiweiss" of the animals. Thus, the metabolic rate should be expressed per N of the organs in which the metabolism is mainly localized. Hereby, it should be possible to eliminate the errors certainly involved in the determinations of metabolism/N and arising from the fact that very often the skeleton of larger animals is relatively heavier (most marked, for example, in the case of many insects).

HOESSLIN (40) emphasized many processes occurring in living organisms to be located in internal or external surfaces of the body (for example, absorbing surfaces of the intestine) and he points (p. 361) at the possibility that the metabolic rate of animals is governed i. a. by the blood supply (and, thus, O_2 supply) to the tissues. For example, the cross sectional area of the aorta of comparable animals is proportional to the surface of the animals. Other surface functions have been mentioned by PÜTTER (93) and others.

It should also be mentioned here that HOESSLIN (40) and ZUNTZ (119) assumed a relation to exist between the activity of the animals and the decrease in metabolic rate with increasing body size. They started from the view that small animals necessarily must be quicker or just as quick as larger animals (i. e. relatively more active) in order to stand the competition. Obviously, this would involve that small animals had a higher metabolic rate than large animals. BUDDENBROCK (18), however, determined the velocity of motion of a large series of animals and he found that the time required by different animals to move over a distance corresponding to their own length is almost the same for small and for large animals. The results of these measurements have been used by BUDDENBROCK as an argument in favour of the view that small animals need relatively less energy for their motion than do large ones. This should explain the unambiguous deviation from the surface law in the case of warm-blooded animals actually found by BENEDICT (5). The present author, however, considers such an explanation unnecessary, since we can hardly give any reason for sticking mentally to the "surface law" which BENEDICT's experiments showed not to be valid.

However, numerous factors tend to make the ratio metabolism/weight of larger animals smaller than that of small animals. It should especially be kept in mind that many physiological processes (absorption from the intestine, osmoregulation, oxygen absorption, heat loss, etc.) are located to surfaces. The processes in question may be energy consuming, they may limit the energy supply of the underlying tissues or govern their energy production. To which extent the surface processes may turn the metabolism of the whole organism from a proportionality with the weight towards a proportionality with the surface must depend on their quantitative importance. At present, we know little or nothing about this problem. Unfortunately, therefore, our discussion is of a qualitative, not of a quantitative nature.

The present author would like to state that he does not consider the factors mentioned in this chapter to be very important. The reasons for this view are to be found in 5.23, but the author just refers to the fact that the magnitude of the decrease in metabolic rate with increasing body size seems to depend on whether we deal with large or with small animals.

Mention should also be made of some recent works which seem to show that the metabolic rate of single organs (tissue slices) decreases with increasing weight of the animals in the same way as does the metabolic rate of the intact animals (KLEIBER (51), KLEIBER, WEYMOUTH and FIELD (52)). ROSENTHAL and DRABKIN (97) found that the cytochrome-C concentration of the kidney cortex decreases with increasing body size of the animals. In trying to explain why the metabolic rate decreases with increasing body weight of the animals we have obviously to consider the chemical composition of the single cells, in casu their enzymatic composition. The whole problem appears to be of a very complex nature and, so far, it can only be raised. As it has been reported in a preceding chapter of this discussion (5.23), the author holds the view that the problem should be considered in relation to the development of the individual. During growth, body size as well as metabolism are governed by genetic and endocrine factors.

5.25. The Mathematical Description of the Phenomena.

It is well known that the ratio between the size of a single organ and that of the whole animal seldom remains constant throughout the development of the animal, because the organ may grow more rapidly or more slowly than the whole animal. PEZARD (89) denoted this phenomenon as "heterogony", CHAMPY (23) called it "disharmonic growth". HUXLEY (43) showed that usually the simple exponential function (which we may express as) $y = k W^e$ described the phenomenon sufficiently accurately. Here, W is the size of the whole animal and y is the size of the respective organ. While k is a simple proportionality factor, the constant e characterizes the growth process in question, in other words, the degree of disharmony. Only in the case $e = 1$ we have harmonic growth. If $e > 1$, the respective organ becomes relatively larger and larger, if $e < 1$, it becomes relatively smaller and smaller as the animal grows.

However, the above formula holds not only for growth processes proper but, moreover, for the comparison of the relative dimensions of small and large adult animals from the same zoological group (for example, *coleoptera*). According to NEEDHAM (85), even the relative augmentation in the organism of one or another chemical compound with increasing body size of the animal may often be described—within the same group of animals—by the above given

simple formula. NEEDHAM denoted the study of the way in which part of the animal (morphologically or chemically defined) varies with the total body size of the animal as "heterauxesis".

In the field of respiration physiology, exactly the same formula has been employed in order to describe mathematically how the respiration measured (y) depends on the body weight (W) of the animal studied. Different workers consider e to be a value which is constant within a species or within a group of animals. This is due to two circumstances, *viz.* on the one hand, the phenomena are often described in a very simple way when using the value e and, on the other hand, a great number of authors have directly participated in the discussion pro or contra the surface law $y = k W^{0.67}$, which means pro or contra a given value of the exponent e , while there has never been objections against the exponential function as such.

However, there is some reason to argue against a continued application of a given mathematical function to the description of phenomena which, in this way, cannot be described satisfactorily. This holds, for example, for the study of the relation between metabolism and the body weight of animals. Until the present, this relation could be described with rather satisfactory accuracy by the equation mentioned above. However, the animals studied in the present work are widely different in size and, very often, the results obtained could not be described by one single value of e and, thus, not by the equation $y = k W^e$. When nevertheless using the term e , we find e generally to decrease as the animals considered become larger. Only in the range of very large animals, e may perhaps again slightly increase. The present author, however, considered it simpler and more correct to say that the percentage decrease in the metabolic rate of the animals varies with the body size of the animals, as it appears from Fig. 48. It should be stressed that Fig. 48 represents the mere analysis of the experimental results, it is no attempt at any interpretation.

The experimental results proper, *viz.* the measurement of the amount of oxygen consumed per unit time and the determination of the nitrogen or the wet weight may be presented to the reader in several ways. In this paper, two alternatives have been considered: One possible way is to plot in a double logarithmic coordinate system the oxygen respired by differently sized animals against their weight or body-N. This very common mode of presentation (adopted, for

instance, by BRODY (14) and by NEEDHAM (85)) is the graphic equivalent of the mathematical expression, *viz.* the above mentioned exponential formula. In this way, we describe the phenomena primarily from the point of view of growth. From the curves obtained, however, the respiration intensities of the animals cannot be read easily, which is a serious drawback. The other possible mode of presentation is to express our results in terms of respiration intensities. Thereby, a closer contact can be established with current physiology and biochemistry. When the oxygen consumed per unit of mass in very different animals is known, the analysis may easily be carried further by determining the concentration (or activity in the case of enzymes) of chemical compounds possibly responsible for differences in respiration intensities. Intensity, activity and concentration are comparable dimensions. In this paper, respiration/weight or respiration/N has, therefore, been plotted against the weight or the body-N of the animals.

5.3. *Evolution, Metabolism and Heat Regulation.*

At different places in the animal kingdom, Figs. 45 and 46 seem to reveal a sudden increase in metabolic intensity appearing when we proceed from one group of animals to another. For example, the arthropods seem generally to have a higher metabolic rate than most other equally large invertebrates studied. Among the vertebrates, mammals and birds seem to have a higher metabolic rate than the equally large reptiles, even when measured at the same body temperature (cf. BENEDICT, 3, 4, 5). These findings give rise to speculations concerning the possible significance of the metabolic intensity for the evolution of the animals. However, the present author has found such considerations not to be very promising and, therefore, the discussion will be confined to one single point:

The probable shift upward of the metabolic intensity at the transition from reptiles to birds and mammals can hardly have been without significance for the acquirement of a heat regulation in these two last mentioned groups of animals. Evidently, the heat production must be of a certain intensity in order that the body temperature can be kept considerably above the temperature of the surroundings. Since the heat loss must be governed essentially by the surface of the respective animal, it will have been of the greatest

significance if already in the poikilotherm ancestors of the homoeotherm animals the basal heat production varied more closely with the body surface than with the body weight. In that case, we can more easily understand the evolution of homoeotherm animals of a widely varying body size and invading the most inaccessible parts of the globe. This holds also if, in the course of further evolution, the relationships originally taken over from poikilotherm animals have been more or less modified, for instance, due to processes of selection.

6. SUMMARY

The present paper deals with the old problems centering around body size and metabolism of the animals. As has been known for a long time, the metabolic rate (oxygen consumption per unit of mass) is usually the more intense, the smaller the animal.

As information into the metabolic rate of micro-animals is highly wanted, the author performed extensive respiration measurements on marine micro-animals. The results together with those available in the literature—the latter, however, mostly obtained on larger animals—should be combined in order to offer a survey of the metabolic rates all over the animal kingdom. However, incomplete this survey will appear, it may be hoped to be of some value in showing the way for further experiments.

Chapter 1 is an introduction to problems treated in this paper.

Chapter 2 describes the technique employed. The Cartesian diver method introduced by LINDERSTRØM-LANG, and further developed by him and his collaborators, is well suited for measuring the metabolism of micro-animals. In the large diver, several very small micro-animals or single somewhat larger ones may be studied. In the small diver, the respiration of single very small micro-animals, or even parts of them, may be determined.

The important and difficult problem of obtaining a measure of the mass of the respective animals was solved by determining their body-N. The N-analyses were performed by means of a micro-method developed recently in the Carlsberg Laboratory. The metabolic rates determined in this way (metabolism/N) are, however,

not comparable with those of larger animals given in the literature, since the mass of these animals is always expressed simply by their body weight. The link connecting the author's experiments with those available in the literature was obtained by performing new measurements on somewhat larger animals. Here, the metabolic rates were measured as metabolism/N and, at the same time, also as metabolism/weight.

In his own experiments the author has disregarded the concept of "standard conditions". Instead, it was attempted to maintain conditions securing "natural activity".

Chapter 3: Experimental part. Since, as already mentioned, the present experiments on animals in "natural activity" have to be compared with experiments performed by other investigators studying resting animals, it is of importance to estimate which fraction of the oxygen uptake of the micro-animals studied results from the work of swimming, crawling, ventilating, etc. Prolonged diver experiments showed the oxygen consumption of many planktonic micro-animals to vary with their activity, which could be observed in the microscope. At the transition from rest to any state of activity the metabolism was usually less than doubled. Experiments on the isolated velum of single lamellibranch larvae, the swimming metabolism of which was known on beforehand, substantiated the following conclusion: The metabolism of the active micro-animal is not very much higher than that of the resting one. Hence, without introducing serious errors, we may compare the metabolic rate of our micro-animals with that of resting animals studied by other workers.

The comparison mentioned is presented in the following chapter.

Some quite different experiments concerning the metabolism of the marine micro-animals should be briefly reported here. 1) The metabolism is independent of the oxygen pressure at least as long as the O_2 -pressure is above 6-7 % of one atmosphere. 2) The temperature influences the metabolism of the micro-animals almost in the same manner as found by KROGH for other animals. 3) Less than 3-8 % of the total oxygen consumed by a lamellibranch larva at 16° C is necessary for keeping the larva float and swim.

Chapter 4. Theoretical part. The experimental results available are presented in graphs showing, for single species or for different important groups of animals, how the metabolic rate of the animals quite generally decreases with increasing body size.

On an average, the micro-fauna respire c. 4-5 times more intensely than the larger marine animals weighing 10^4 - 10^5 times more. The same result is obtained irrespective of whether we compare larvae and adults of one species, or we compare small and large related species.

By comparing in one and the same figure all average curves described above, we gain an, admittedly very rough and very incomplete, survey of the respiration intensities throughout the animal kingdom. This survey is represented in Figs. 45 and 46 of this paper. Most experiments on cold-blooded animals have been performed at temperatures around 16° C. Warm-blooded animals, of course, had their normal body temperature.

It is considered important that the percentage decrease in metabolic rate found for a certain increase in body size is not a constant value, nor do the observations scatter statistically around a mean value. The percentage decrease depends clearly upon whether we consider small or large animals, and is the smaller, the smaller the animals. This appears from the analysis given in Fig. 48 of the slopes of the curves illustrating the metabolic rate-body size relations.

Chapter 5: Discussion. The just mentioned Fig. 48 seems to indicate a strange connection to be present throughout the animal kingdom, a connection which, indeed, is difficult to understand and, hence, also difficult to accept. The argumentation given leads to the conclusion that Fig. 48, in fact, results from comparing experiments which are not quite comparable. So far, no distinction has been made—but in the future this will be necessary—between comparisons of animals belonging to different species and comparisons of animals which belong to a single species. In the latter case, we should especially distinguish between comparisons of animals in which growth is about to stop and comparisons of animals which are still growing intensely.

In fact, the tendency revealed in Fig. 48 may be explained simply by assuming the metabolic rate to change in a typical manner as the individual develops from egg to adult animal: During the main period of growth the percentage decrease in metabolic rate becomes more and more pronounced; an especially steep decrease sets in when the individual is approaching its final body size, i. e. probably at the time when growth is about to stop. Hence, according to this view, steep de-

creases in the metabolic rate are found in species of very different sizes.

Experimental evidence is in favour of the assumed typical change in metabolic rate during the development of the individual. If this view is correct, the inhomogeneity of the experimental material underlying Fig. 48 explains rather well the tendency revealed in the figure.

To the author's opinion, the assumed change in metabolic rate during the life of the individual is in itself of considerable interest from a biochemical point of view.

7. REFERENCES

1. ALLEN, GEORGE D.: *Am. J. Physiol.* **49**, 420—473. 1919.
2. BARRON, DONALD H.: *Developmental Physiology*, *Ann. Rev. of Physiol.* **7**, 107—126, 1945.
3. BENEDICT, F. G.: *The Physiology of Large Reptiles*. Carnegie Publ. **425**. Washington 1932.
4. BENEDICT, F. G.: *Hibernation and Marmot Physiology*. Carnegie Publ. **497**. Washington 1938.
5. BENEDICT, F. G.: *Vital Energetics*. Carnegie Publ. **503**. Washington 1938.
6. BLANK, H.: *Arch. f. d. ges. Physiol.* **234**, 310—317. 1934.
7. BODINE, J. H.: *J. exper. Zool.* **32**, 137—164, 1921.
8. BODINE, J. H. and T. C. EVANS: *Biol. Bull.* **63**, 235. 1932.
9. BOELL, E. J. and J. S. NICHOLAS: *Science* **90**, 411. 1939.
10. BOELL, E. J.: *Trans. Connecticut Acad. Art and Sci.* **36**, 429—441. 1945.
11. BORNEBUSCH, C. H.: *The Fauna of Forest Soil*. (Diss.). Copenhagen. 1930.
12. BOUNHIOL, JEAN: *Ann. d. Sci. natur. Zool. Sér. 8*, **16**, 1—132. 1902.
13. BRACHET, J.: *Embryologie Chimique*, Paris-Liège, 1945.
14. BRODY, SAMUEL: *Bioenergetics and Growth*, New York, 1945.
15. BRÜEL, D., HEINZ HOLTER, K. LINDERSTRØM-LANG and K. ROZITS: *Compt. rend. Lab. Carlsberg, Sér. Chim.* **25**, 289—323. 1946.
16. BRUCE, JOHN R.: *Bioch. Journ.* **20**, 829—846. 1926.
17. BUDDENBROCK, W. v., und G. v. ROHR: *Arch. f. d. ges. Physiol.* **194**, 468—472. 1922.
18. BUDDENBROCK, W. v.: *Naturw.* **22**, 675—680. 1934.
19. BUDDENBROCK, W. v.: *Grundriss der vergleichenden Physiologie*, Bd. 2. Berlin 1939.

20. BUTLER, R. G. and J. M. INNES: Proc. Roy. Soc., London s. B. **119**, 296—304. 1936.
21. BUYTENDIJK, F. J. J.: Kon. Akad. van Wetensch. Amsterdam. Proc. Sect. of Sci. **12**, 48—54. 1910.
22. BUYTENDIJK, F. J. J.: Kon. Akad. van Wetensch. Amsterdam. **B. I**, 44—53. 1910.
23. CHAMPY, C.: Sexualité et Hormones. Paris 1924.
24. COHNHEIM, O.: Z. f. physiol. Chem. **76**, 298—313. 1912.
25. DOUDOROFF, P.: Biol. Bull. **83**, 219—244. 1942.
26. EDWARDS, G. A. and LAURENCE IRVING: Journ. cell. comp. Physiol. **21**, 169—182. 1943.
27. FOX, H. MUNRO: Proc. Zool. Soc., London part. 2, 945—955. 1936. Ibid. s. A, **108**, 501—505. 1938.
28. FOX, H. MUNRO: Proc. Zool. Soc. London s. A., **109**. 1939.
29. FOX, H. MUNRO and B. G. SIMMONDS: Journ. exper. Biol. **10**, 67. 1933.
30. GOMPEL, M.: Ann. de Physiol. **14**, 914—931. 1938.
31. GRAY, J.: Ciliary Movement. Cambridge, 1928.
32. GROEBBELS, FRANZ: Z. f. Biol. **75**, 155—168. 1922.
33. GROEBBELS, FRANZ: Arch. f. d. ges. Physiol. **208**, 718—729. 1925.
34. GROEBBELS, FRANZ: Der Vogel I, Atmungswelt und Nahrungswelt. Berlin 1932.
35. GUNN, D. L.: J. exper. Biol. **12**, 185. 1935.
36. HALL, V. E.: Biol. Bull. **61**, 400—416. 1931.
37. HARNISCH, OTTO: Z. f. vergl. Physiol. **22**, 450—465. 1935.
38. HAUGAARD, NIELS and LAURENCE IRVING: Journ. cell. comp. Physiol. **21**, 19—26. 1943.
39. HENZE, MARTIN: Bioch. Z. **26**, 255—278. 1910.
40. HOESSLIN, H. v.: Arch. f. Anat. Physiol., Physiol. Abt. 323—379. 1888.
41. HOLTER, H.: Compt. rend. Lab. Carlsberg, Sér. Chim. **24**, 399—478. 1943.
42. HOLTER, HEINZ and ERIK ZEUTHEN: Compt. rend. Lab. Carlsberg, Sér. Chim. **25**, 33—65. 1944.
43. HUXLEY, JULIAN S.: Problems of Relative Growth. London 1932.
44. HYMAN, L. H.: Am. J. Physiol. **50**, 67—81. 1919.
45. JENSEN, PAUL: Arch. f. d. ges. Physiol. **54**, 538—551. 1893.
46. JOLYET, F. et PAUL REGNARD: Arch. de Physiol. 2. Sér. **4**, 584. 1877.
47. JØRGENSEN, C. BARKER: Acta Physiol. Scand. **5**, 297—304. 1943.
48. KESTNER, O.: Arch. f. d. ges. Physiol. **234**, 290—301. 1934.
49. KITTEL, ANNEISE: Z. f. vergl. Physiol. **28**, 533—562. 1941.
50. KLEIBER, MAX: Energy Metabolism. Ann. Rev. of Physiol. **VI**, 123—154. 1944.
51. KLEIBER, MAX: Pröc. Soc. Exper. Biol. and Med. **48**, 419. 1941.
52. KLEIBER, M., WEYMOUTH, F. W. and FIELD, J. II: Ibid. **49**, 367. 1942.
53. KNAUTHE, KARL: Arch. f. d. ges. Physiol. **73**, 490—500. 1898.
54. KONOPACKI, M.: Mem. Acad. Sc. Cracovie. 357. 1907.
55. KRAMER, GUSTAV: Z. f. vergl. Physiol. **20**, 600—616. 1934.
56. KRAMER, GUSTAV: Z. f. vergl. Physiol. **22**, 39—49. 1935.

57. KREHL, L. and F. SOETBEER: Arch. f. d. ges. Physiol. **77**, 611—638. 1899.
58. KROGH, AUG.: Bioch. Z. **62**, 266—279. 1914. *ibid.* **66**, 512. 1914.
59. KROGH, AUG.: Internat. Z. f. physik.-chem. Biol. **1**, 491—508. 1914.
60. KROGH, AUG.: The Respiratory Exchange of Animals and Man. London 1916.
61. KROGH, AUG.: Biol. Rev. **VI**, 412—442. 1931.
62. KROGH, AUG. and ANCEL KEYS: Biol. Bull. **67**, 132—144. 1934.
63. KROGH, AUG. and R. SPÄRCK: Kgl. Danske Vidensk. Selsk., Biol. Medd. **13**, Nr. 4. 1936.
64. KROGH, AUG.: Industr. and Engin. Chem. **7**, 131—133. 1935.
65. KRÜGER, F.: Z. wiss. Zool. **152**, 547—570. 1940.
66. KUHN, OSCAR: Die fossilen Reptilen. Berlin 1937.
67. KUHN, OSCAR: Die Phylogenie der Wirbelthiere auf paläontologischer Grundlage. Jena 1938.
68. LANG, JAROSLAV: Biol. Zbl. **54**, 85—88. 1934.
69. LEVY, MILTON: Compt. rend. Lab. Carlsberg, Sér. Chim. **21**, 101—110. 1936.
70. LIANG, CHUNG MOU: Arch. f. d. ges. Physiol. **234**, 302—309. 1934.
71. LIEBSCH, W.: Zool. Jahrb. Abt. allg. Zool. Physiol. **46**, 161—208. 1929.
72. LINDERSTRØM-LANG, K.: Nature. **140**, 108. 1937.
73. LINDERSTRØM-LANG, K.: Compt. rend. Lab. Carlsberg, Sér. Chim. **24**, 249—280. 1942.
74. LINDERSTRØM-LANG, K. and H. HOLTER: Compt. rend. Lab. Carlsberg, Sér. Chim. **24**, 105—138. 1942.
75. LINDERSTRØM-LANG, K.: Compt. rend. Lab. Carlsberg, Sér. Chim. **24**, 333—398. 1943.
76. LINDROTH, ARNE: Zool. Bidrag från Uppsala. (Diss.). **17**, 367—497. 1938.
77. LUDWIG, WILHELM: Z. f. vergl. Physiol. **24**, 319—342. 1937.
78. LUDWIG, WILHELM: Z. f. vergl. Physiol. **13**, 398—504. 1931.
79. LUND, E. J.: Biol. Bull. **41**, 203—220. 1921.
80. MARSHALL, S. M. and A. G. NICHOLLS: Journ. Mar. Biol. Ass. **20**, 1—27. 1935—36.
81. MEYER, HELGA: Zool. Jahrb. Abt. allg. Zool. Physiol. **55**, 349—398. 1935.
82. MICHAL, K.: Zool. Anz. **95**, 65—75. 1931.
83. MONTUORI, A.: Arch. ital. Biol. **59**, 213—234. 1913.
84. MORGULIS, S.: Am. J. Physiol. **36**, 207—216. 1915.
85. NEEDHAM, JOSEPH: Biochemistry and Morphogenesis. Cambridge 1942.
86. OBRESHKOVE, V. and A. M. BANTA: Physiol. Zool. **3**, 1930.
87. PANTIN, C. F. A.: Journ. exp. Biol. **8**, 63 and 82. 1931.
88. PARNAS, J. K.: Z. für analyt. Chem. **114**, 261—275. 1938.
89. PÉZARD: cit. from NEEDHAM (85), p. 533.
90. PINCUS: The Egg of Mammals, New York, 1936.
91. PHILIPS, F. S.: Journ. exper. Zool., **86**, 257. 1941. *ibid.* **90**, 83. 1942.
92. POTT, R.: Landwirtsch. Versuchsstationen. **18**, 81. 1875.

93. PÜTTER, AUG.: Die Ernährung der Wassertiere. Jena 1909.
94. PÜTTER, AUG.: Z. allg. Physiol. **6**, 217—286. 1907. *ibid.* **7**, 16—61. 1908.
95. RAFFY, ANNE: Compt. rend. Acad. de sci. **196**, 374—376. 1933.
96. REGNAULT, V. et I. REISET: Ann. de Chim. et de Phys. Sér. 3, **26**, 299—520. 1849.
97. ROSENTHAL, O. and D. L. DRABKIN: Journ. Biol. Chem. **150**, 131—141. 1943.
98. RUBNER, MAX: Z. für Biol. **19**, 535—562. 1883.
99. RUBNER, MAX: Biologische Gesetze. Marburg 1887.
100. RUBNER, MAX: Sitz.ber. der preuss. Akad. Wiss. 217—234. 1924.
101. SPÄRCK, R.: Rep. dansk Biol. Stat. **35**, 103—105. 1929.
102. SPÄRCK, R.: XI Congr. intern. Zool., Padova. Vol. II, 900—904. 1930.
103. SPÄRCK, R.: Kgl. Danske Vidensk. Selsk., Biol. Medd. **13**, Nr. 5. 1936.
104. TEISSIER, GEORGES: Ann. de Physiol. **4**, 1—26. 1928.
105. TEISSIER, GEORGES: Trav. stat. biologique Roscoff. **9**, 27—238. 1931.
106. TERROINE, E. F. et G. DELPECH: Ann. de Physiol. **7**, 341—381. 1931.
107. THAMDRUP, H. M.: Medd. Komm. Danm. Fiskeri- og Havunders. Ser. Fiskeri. (Diss.). **10**, 2. 1935.
108. THORSON, GUNNAR: Medd. om Grønland. (Diss.). **100**. Copenhagen 1936.
109. THORSON, GUNNAR: Reproduction and larval development of danish marine invertebrates. Medd. Komm. Danm. Fiskeri- og Havunders. Ser. Plankton. **4**. 1946.
110. THUNBERG, THORSTEN: Skandinav. Arch. f. Physiol. **17**, 133—195. 1905.
111. VERNON, B. A.: Journ. Physiol. **19**, 18—70. 1895—96.
112. WEINLAND, E.: Z. für Biol. **69**, 1—86. 1919.
113. WELLS, N. A.: Biol. Bull.: **69**, 361. 1935.
114. WEYMOUTH, F. W., J. M. CRISMON, V. E. HALL, H. S. BELDING and JOHN FIELD II: Physiol. Zoöl. **17**, 50. 1944.
115. WHITNEY, R. J.: Journ. exper. Biol. **19**, 168—175. 1942.
116. WINDLE, W. F.: Physiology of the Fetus. Philadelphia and London 1940.
117. WINGFIELD, C. A.: Proc. Zool. Soc. London. A. **109**. 1939.
118. ZEUTHEN, E.: Compt. rend. Lab. Carlsberg, Sér. Chim. **24**, 479—518. 1943.
119. ZUNTZ, N.: Nachrichten aus d. Klub d. Landwirte zu Berlin. No. 448, 4480. 1906. (cit. from KROGH (60)).

DANSK OVERSIGT

Denne Afhandling behandler det gamle Problem om Forholdet mellem Dyrenes Størrelse og deres Stofskifte (Iltoptagelse). Det har længe været kendt, at jo mindre et Dyr er, desto mere intenst forløber Omsætningerne i dets Væv, desto højere Iltforbrug har hvert Gram af Dyret.

Siden KROGH's Sammenfatning fra 1916 er vor Viden om Dyrenes Iltforbrug meget væsentlig forøget, dog er de mellemstore og store Dyr stadig stærkest repræsenteret i Literaturen. Stofskiftet af de Dyr, hvis Vægt maales i mg eller i γ er næsten eller helt ukendt, og Forfatteren har set det som sin experimentelle Opgave at fremskaffe brugbare Stofskiftemaalinger ogsaa paa disse meget smaa Dyr. Øresunds marine Mikrofauna er derfor indgaaende studeret med Anvendelse af Carlsberg Laboratoriets Mikrometoder. De saaledes vundne Maalinger i Forbindelse med det i Literaturen nedfældede Forsøgsmateriale giver et saa fyldigt Udsnit af Dyreriget, baade hvad angaar Arter, Dyregrupper og Størrelsesvariation af de studerede Dyr, at en Syntese tør forsøges. Naar man saaledes overskuer hele Dyreriget, maa Resultatet ifølge Sagens Natur altid forblive yderst ufuldkomment, og det kan i bedste Fald vise frem til nye Maader at betragte Problemerne paa.

Kapitel 1 i denne Afhandling er en Indledning.

Kapitel 2 omhandler den anvendte Teknik samt visse biologiske Forhold ved Maalingerne. LINDERSTRØM-LANG's Cartesianske Dykker i HOLTER's Udformning er velegnet til Maaling af Iltforbrug i Størrelsesordenen 10^{-1} - 10^{-2} μ l/Time. I denne Dykker kan Stofskiftet (ved 16° C) af et begrænset Antal mindre Mikrodyr (Længde 0.1-0.3 mm) studeres, eller man kan maale Stofskiftet af det enkelte, lidt større Mikrodyr. I den Dykker, der er udformet af ZEUTHEN og som er 10-20 Gange følsommere end HOLTER's Dykker, kan et enkelt af de ganske smaa Dyr studeres, omend med lidt større Besvær.

Det vigtige og vanskelige Punkt, at faa et Maal for Mængden af det Væv, der har været benyttet til hvert Aandingsforsøg, løstes ved (med Modifikationer) at anvende Carlsberg-Metoden til N-Bestemmelse (BRÜEL, HOLTER, LINDERSTRØM-LANG og ROZITS). Denne Metode er netop beregnet til Bestemmelse af N i de smaa Mængder, der normalt forefindes i de Væv, der tages til Forsøg i Dykkerne.

Kvælstoffet i de Dyr, som studeredes i ZEUTHEN's Dykker, var dog for sparsomt til at kunne bestemmes direkte, men dets Mængde kunde ofte skønnes ad indirekte Vej.

Forholdet $\frac{\text{Stofskifte}}{\text{Legems-N}}$ betegnes i denne Afhandling som Dyrets Stofskifteintensitet (metabolic rate, respiratory rate). Forfatterens Forsøg er imidlertid ikke direkte sammenlignelige med Forsøg fra Literaturen udført paa større Dyr, fordi man altid tidligere har maalt Stofskifteintensiteten ved Forholdet $\frac{\text{Stofskifte}}{\text{Legemsvægt}}$. Det blev derfor nødvendigt, med lidt grovere Metoder, at udføre nye Forsøg, i hvilke Stofskifteintensiteten hos de noget større Havdyr samtidig bestemtes paa begge de ovennævnte Maader. Disse Forsøg repræsenterer saaledes det Mellemlid, der gør nærværende Mikroforsøg sammenlignelige med de talrige Forsøg, der af andre Forfattere er udført paa større og store Dyr.

Der er i denne Afhandling helt set bort fra Kravet om Standardbetingelser. Alle Forsøg er udført ved 16° C, men med Dyr i saavidt muligt "naturlig Aktivitet".

Kapitel 3: Experimentel Del. Da egne Forsøg, der er udført paa Mikrodyr i "naturlig Aktivitet" senere skal sammenlignes med andre Forsøg paa større hvilende Dyr, maa vi forsøge at skaffe os Viden om, hvor stor en Del af de smaa Dyrs Stofskifte, der skyldes deres Aktivitet. Til dette Formaål udførtes i flere Tilfælde langstrakte Dykkerforsøg paa et enkelt Mikrodyr fra Planktonet. Samtidig med, at Svingningerne i Dyrets Stofskifte kunde maales, observeredes dets vilkaarlige Aktivitetsændringer i Mikroskopet. Resultatet blev, at naar Dyret skiftede fra Hvile til Svømning eller anden livlig Aktivitet, steg Stofskiftet, dog ikke til mere end det dobbelte af Hvileværdien, ofte mindre. Forsøg, hvor Stofskiftet af en svømmende Muslingelarve sammenlignedes med Stofskiftet af samme Dyrs isolerede, livligt svømmende Fimresejl, viste bekræftende, at højst Halvdelen af Dyrets Stofskifte var lokaliseret til Bevægelsesorganet (*Mytilus* 50 %, *Teredinide* 15 %). Disse Resultater indebærer den vigtige Konsekvens, at man rimeligvis uden at begaa meget væsentlige Fejl, kan sammenligne Forsøg paa Mikrodyr i "naturlig Aktivitet" med de i Literaturen meddelte Forsøg paa større, hvilende Dyr.

Selve Bestemmelserne af Dyrenes Stofskifteintensiteter meddeles i Kapitel 4.

Af specielle Forhold ved Mikrofaunaens Stofskifte skal nævnes:

Stofskiftet viste sig uafhængig af Iltrykket ned til — og maaske under — et Iltryk svarende til 6—7 % af en Atm. Temperaturen's Indflydelse paa Mikrodyrenes Aanding er omtrent den samme som udtrykt i KROGH's Standardkurve. Et ganske ringe Energiforbrug ($< 3-8\%$ af det samlede Energiforbrug ved 16°C) er tilstrækkeligt til at en Muslingelarve kan svæve og svømme i Planktonet.

Kapitel 4: Teoretisk Del. Inden for de vigtigste Dyregrupper afbildes grafisk — med Anvendelse af egne og andres Forsøg — Sammenhængen mellem Dyrenes Aandingsintensitet og deres Størrelse (N, Vægt). Det bliver klart, at Dyrenes Stofskifteintensitet ganske alment falder med deres stigende Størrelse. Gennemsnitlig aander Mikrofaunaen 4-5 Gange saa intenst som de større Havdyr, hvis Masse er 10^4-10^5 Gange saa stor. Det er herved ligegyldigt, om man sammenligner Larver med voksne Former af samme Art, eller om smaa og store Arter sammenlignes.

Ved Sammenligning i een Figur (se Fig. 45 og 46) af alle Gennemsnitskurver, der beskriver Sammenhængen mellem Størrelse og Stofskifte for de enkelte Arter eller Dyregrupper, skaffes et groft Overblik over Stofskifteintensiteterne i Dyreriget. For de koldblodede Dyrs Vedkommende har Forsøgstemperaturen været nær 16°C ; de varmblodede Dyrs Stofskifte er naturligvis maalt ved almindelig Legemstemperatur.

Det er muligt, at der ved Overgang fra een Dyregruppe til en anden kan indtræde Spring i Dyrenes gennemsnitlige Stofskifteintensitet.

Undersøges den Hastighed, hvormed Dyrenes Stofskifteintensitet falder med deres stigende Størrelse, ses, at denne Hastighed er afhængig af, om man har at gøre med smaa eller med store Dyr. Det procentiske Fald i Stofskifte ved en vis Forøgelse af de betragtede Dyrs Størrelse er mindre for smaa end for større Dyr (se Fig. 48).

Kapitel 5: Diskussion. Der synes gennem den lige nævnte Fig. 48 at afsløres en indre Sammenhæng i hele Dyreriget, en Sammenhæng, der dog er yderst svær at forstaa og dermed ogsaa svær at acceptere, og der argumenteres da ogsaa for, at Fig. 48 i Virkeligheden er fremkommet ved Sammenligning af ikke fuldt sammenlignelige Forsøg. Man maa f. Ex. skelne mellem Sammenligninger af forskelligt store Dyr af forskellig Art og Sammenligninger af forskelligt store Dyr af samme Art. For de sidstnævnte Dyrs Vedkommende synes mange Forsøg i Literaturen at tyde paa, at der skarpt

maa skelnes mellem Sammenligninger af Dyr, der endnu er i kraftig Vækst og Sammenligninger af Dyr, der nærmer sig sin størst mulige Størrelse, d. v. s. hvis Vækst er stærkt bremset.

Faktisk kan man forklare, at Observationerne i Fig. 48 fordeler sig skævt, ved Antagelsen, at Stofskiftets Intensitet altid gennem det enkelte Individets Udvikling fra Æg til udvokset Dyr varierer paa typisk Maade: Under hele Væksten falder Stofskifteintensiteten procentisk stærkere og stærkere, men et særligt stærkt Fald indtræder, naar Individet nærmer sig sin endelige Størrelse, det vil maaske sige, naar Væksten (paa Grund af genetiske og endokrine Faktorer) gaar i Staa. Hvis Stofskifteintensitetens Gang gennem det enkelte Dyrs Liv virkelig i Hovedsagen svarer til det skitserede — og der synes at være stærk experimentel Støtte for en saadan Antagelse — vil alene Heterogeniteten i det Forsøgsmateriale, der ligger til Grund for Fig. 48, forklare Figurens Tendens. Selve den skitserede sandsynlige Ændring i Stofskifteintensiteten gennem det enkelte Dyrs Ontogenese forekommer Forfatteren at have betydelig fysiologisk og biokemisk Interesse.

BIANCO LUNOS BOGTRYKKERI